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BIOASSAY OF 1,5-NAPHTHALENEDIAMINE FOR POSSIBLE CARCINOGENICITY

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BIOASSAY OF

1,5-NAPHTHALENEDIAMINE

FOR POSSIBLE CARCINOGENICITY

Carcinogenesis Testing Program
Division of Cancer Cause and Prevention
U.S. National Cancer Institute
″ National Institutes of Health
Bethesda, Maryland 20014

Carcinogenesis Technical report series

U.S. DEPARTMENT OF HEALTH, EDUCATION, AND WELFARE
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REPORT ON THE BIOASSAY OF 1,5-NAPHTHALENEDIAMINE FOR POSSIBLE CARCINOGENICITY

CARCINOGENESIS TESTING PROGRAM
DIVISION OF CANCER CAUSE AND PREVENTION
NATIONAL CANCER INSTITUTE, NATIONAL INSTITUTES OF HEALTH

FOREWORD: This report presents the results of the bioassay of 1,5-naphthalenediamine conducted for the Carcinogenesis Testing Program, Division of Cancer Cause and Prevention, National Cancer Institute (NCI), National Institutes of Health, Bethesda, Maryland. This is one of a series of experiments designed to determine whether selected chemicals have the capacity to produce cancer in animals. Negative results, in which the test animals do not have a significantly greater incidence of cancer than control animals, do not necessarily mean the test chemical is not a carcinogen because the experiments are conducted under a limited set of circumstances. Positive results demonstrate that the test chemical is carcinogenic for animals under the conditions of the test and indicate a potential risk to man. The actual determination of the risk to man from animal carcinogens requires a wider analysis.

CONTRIBUTORS: This bioassay of 1,5-naphthalenediamine was conducted by Mason Research Institute, Worcester, Massachusetts, initially under direct contract to the NCI and currently under a subcontract to Tracor Jitco, Inc., prime contractor for the NCI Carcinogenesis Testing Program.

The experimental design was determined by the NCI Project Officers, Dr. J. H. Weisburger (1,2) and Dr. E. K. Weisburger (1). The principal investigators for the contract were Dr. E. Smith (3) and Dr. A. Handler (3). Animal treatment and observation were supervised by Mr. G. Wade (3) and Ms. E. Zepp (3). Chemical analysis was performed by Midwest Research Institute (4) and the analytical results were reviewed by Dr. N. Zimmerman (5).

Histopathologic examinations were performed by Dr. A. S. Krishna Murthy (3), Dr. A. Russfield (3) and Dr. D. S. Wyand (3) at the Mason Research Institute, the pathology narratives were written by Dr. A. Russfield (3) and Dr. D. S. Wyand (3), and the diagnoses included in this report represent the interpretation of these pathologists. Histopathology findings and reports were reviewed by Dr. R. L. Schueler (6).

Compilation of individual animal survival, pathology, and summary tables was performed by EG&G Mason Research Institute (7); the statistical analysis was performed by Mr. W. W. Belew (5,8) and Mr. R. M. Helfand (5), using methods selected for the Carcinogenesis Testing Program by Dr. J. J. Gart (9).

This report was prepared at METREK, a Division of The MITRE Corporation (5) under the direction of the NCI. Those responsible for this report at METREK are the project coordinator, Dr. L. W. Thomas (5), task leader Dr. M. R. Kornreich (5,10), senior biologist Ms. P. Walker (5), biochemist Mr. S. C. Drill (5), and technical editor Ms. P. A. Miller (5). The final report was reviewed by members of the participating organizations.

The following other scientists at the National Cancer Institute were responsible for evaluating the bioassay experiment, interpreting the results, and reporting the findings: Dr. K. C. Chu (1), Dr. C. Cueto, Jr. (1), Dr. J. F. Douglas (1), Dr. D. G. Goodman (1,10), Dr. R. A. Griesemer (1), Dr. M. H. Levitt (1), Dr. H. A. Milman (1), Dr. T. W. Orme (1), Dr. R. A. Squire (1,11), Dr. S. F. Stinson (1), Dr. J. M. Ward (1), and Dr. C. E. Whitmire (1).

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SUMMARY

A bioassay of 1,5-naphthalenediamine for possible carcinogenicity was conducted using Fischer 344 rats and B6C3F1 mice. 1,5-Naphthalenediamine was administered in the feed, at either of two concentrations, to groups of 50 male and 50 female animals of each species. The high and low dietary concentrations utilized in the chronic bioassay were, respectively, 0.1 and 0.05 percent for rats and 0.2 and 0.1 percent for mice. The compound was administered in the diet for 103 weeks, followed by up to 4 weeks of observation. Fifty mice of each sex and 25 rats of each sex were placed on test as controls. These animals were observed for up to 110 weeks.

There were no significant positive associations between the administered concentrations of 1,5-naphthalenediamine and mortality in either sex of rats or mice. In all groups adequate numbers of animals survived sufficiently long to be at risk from late-developing tumors.

Among dosed female rats, a statistically significant increase in endometrial stromal polyps was observed. Several of these tumors underwent malignant transformation to endometrial stromal sarcomas. The incidence of female rats having either adenoma or carcinoma of the clitoral gland was statistically significant. No neoplasms were observed at significantly increased incidences in dosed male rats. Based on lack of clinical signs or weight loss, the male rats may have been able to withstand a higher dose.

In mice, dose-related increases in thyroid neoplasms were observed in both sexes. The incidence of thyroid C-cell carcinomas was significant for high dose female mice. The combined incidences of papillary adenomas, follicular-cell adenomas and papillary cystadenomas of the thyroid were significant for mice of both sexes. The incidence of hepatocellular carcinomas and the incidence of alveolar/bronchiolar adenomas were each significant for dosed female mice.

Under the conditions of this bioassay, 1,5-naphthalenediamine was carcinogenic in female Fischer 344 rats, causing clitoral and uterine neoplasms. 1,5-Naphthalenediamine was also carcinogenic for B6C3F1 mice, producing thyroid neoplasms in males and neoplasms of the thyroid, liver, and lung in females. Insufficient evidence was provided for the carcinogenicity of the compound in male Fischer 344 rats.

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I. INTRODUCTION

1,5-Naphthalenediamine (Figure 1) (NCI No. C03021), a bicyclic aromatic amine used in the dye industry, was selected for bioassay by the National Cancer Institute because of the high incidence of bladder cancer reported among dye manufacturing industry workers (Anthony and Thomas, 1970; Wynder et al., 1963). Aromatic amines are one class of chemicals believed to contribute to the increased cancer risk in this industry (Wynder et al., 1963). The structural similarity of 1,5-naphthalenediamine to both the human bladder carcinogen 2-naphthylamine (International Agency for Research on Cancer [IARC], 1974) and the suspected carcinogen 1-naphthylamine (IARC, 1974) was an additional factor in its selection for testing.

The Chemical Abstracts Service (CAS) Ninth Collective Index (1977) name for this compound is 1,5-naphthalenediamine.* It is also known as 1,5-diaminonaphthalene.

1,5-Naphthalenediamine can be used as an oxidation base (Colour Index [C.I.] 76595), an intermediate in the synthesis of the dye Naphthylene Red (C.I. 21650) (Society of Dyers and Colourists, 1956), and in the production of a black trisazo dye for cotton (Taube, 1973). 1,5-Naphthalenediamine has also been used as a precursor for 1,5-naphthalenediisocyanate (Hirai and Yamamoto, 1975); as an intermediate in the synthesis of drugs for the symptomatic treatment of asthma or

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*The CAS registry number is 2243-62-1

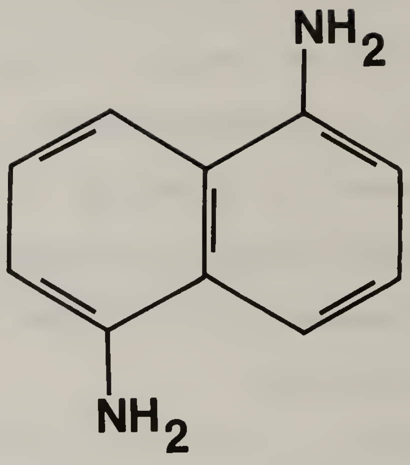

FIGURE 1
CHEMICAL STRUCTURE OF 1,5-NAPHTHALENEDIAMINE

rhinitis (Hall, 1976); as a component of piperazine-modified aromatic polyamides (Fujiwara et al., 1974); and as a modifier for phenolic resins used in rapid curing compounds (Freeman et al., 1974); however, these uses appear to be purely experimental.

Specific production data for 1,5-naphthalenediamine are not available; however, the exclusion of this compound from the 1977 Directory of Chemical Producers, U.S.A. (Stanford Research Institute, 1977) implies that it is not produced in commercial quantities (in excess of 1000 pounds or $1000 in value annually).

The potential for exposure to 1,5-naphthalenediamine may be greatest for workers in the dye industry and persons engaged in chemical research with this compound.

II. MATERIALS AND METHODS

A. Chemicals

1,5-Naphthalenediamine was purchased from Carroll Products, Wood River Junction, Rhode Island by the NCI for Mason Research Institute, Worcester, Massachusetts, and chemical analysis was performed by Midwest Research Institute, Kansas City, Missouri. The experimentally determined melting point of 190° to 191°C suggested a compound of high purity based on its narrow range and its close proximity to the value (190°C) reported in the literature (Pollock and Stevens, 1965). Elemental analysis was consistent with $C_{10}H_{10}N_2$, the molecular formula for 1,5-naphthalenediamine. However, nonaqueous amine group titration was approximately 89 to 90 percent of that expected on a theoretical basis. Vapor-phase chromatography revealed one homogeneous peak, but thin-layer chromatography utilizing two solvent systems (acetone:ammonium hydroxide and methylethylketone:formic acid), each visualized with 254 nm and 367 nm light, indicated the presence of one nonmotile impurity. Nuclear magnetic and infrared analyses were consistent with the structure of the compound. Ultraviolet analysis showed λ_{max} at 232, 328 and 498 nm with ϵ values of 62,800, 10,640 and 9, respectively. The literature (Sadtler Standard Spectra) indicates a λ_{max} at 328.5 nm with ϵ = 10,000 for 1,5-naphthalenediamine. The observed ϵ at 328 nm was 10,640 (6 percent greater than expected).

Throughout this report the term 1,5-naphthalenediamine is used to represent this compound.

B. Dietary Preparation

The basal laboratory diet for both dosed and control animals consisted of Wayne Lab-Blox® (Allied Mills, Inc., Chicago, Illinois). 1,5-Naphthalenediamine was administered to the dosed animals as a component of the diet. Under an exhaust hood, proper amounts of the chemical were removed from the stock bottle. The compound was blended in an aluminum bowl with an aliquot of the ground feed. Once visual homogeneity was attained, the mixture was placed into a 6 kg capacity Patterson-Kelley twin-shell stainless steel V-blender, along with the remainder of the meal and blended for 20 minutes. Prepared diets were placed in double plastic bags and stored in the dark at 4°C. The mixture was used for 1 week only.

C. Animals

Two animal species, rats and mice, were used in the carcinogenicity bioassay. Fischer 344 rats and B6C3F1 mice were obtained through contracts of the Division of Cancer Treatment, National Cancer Institute. All animals were obtained from Charles River Breeding Laboratories, Inc., Wilmington, Massachusetts. Dosed and control animals were received in separate shipments. Upon arrival, a sample of animals was examined for parasites and other signs of disease. All animals appeared to have parasites. They were treated with 3.0 gm of piperazine adipate per liter of drinking water, *ad libitum*, for 3 days, followed by 3 days of plain tapwater and 3 subsequent days of piperazine adipate administration. During this period, new cages with fresh bedding were provided daily. Animals were held in quarantine by species for 2 weeks prior to initiation of test. Animals were assigned to groups and distributed among cages so that average body weight per cage was approximately equal for a given sex and species.

D. Animal Maintenance

All animals were housed by species in rooms having a temperature range of 23° to 34°C. Incoming air was filtered through Tri-Dek® 15/40 denier Dacron® filters (Tri-Dim Filter Corp., Hawthorne, New Jersey) providing six changes of room air per hour. Fluorescent lighting was provided on a 12-hour-daily cycle.

Rats were housed five per cage by sex. During quarantine and for the first 14 months of study rats were housed in galvanized-steel wire-mesh cages suspended over newspapers. Newspapers under cages were replaced daily and cages and racks washed weekly. For the remainder of the study, rats were held in suspended polycarbonate cages equipped with disposable nonwoven fiber filter sheets. Clean bedding and cages were provided twice weekly. SAN-I-CEL® corncob bedding (Paxton Processing Company, Paxton, Illinois) was used for the first 2 months that rats were housed in polycarbonate cages. For the remainder of the study, Aspen hardwood chip bedding (American Excelsior Company, Baltimore, Maryland) was used. Stainless steel cage racks were cleaned once every 2 weeks, and disposable filters were replaced at that time.

Mice were housed by sex in polycarbonate shoe box type cages. Cages were fitted with perforated stainless steel lids (Lab Products, Inc., Garfield, New Jersey). Nonwoven fiber filter bonnets were used over cage lids. Control mice were housed ten per cage for the first month of study and five per cage thereafter. Dosed mice were held five per cage throughout the study. Clean cages, lids, and bedding were provided twice per week. SAN-I-CEL® was used during the first 9 months of study. A second corncob bedding (Bed-o-Cobs®, The Andersons Cob Division, Maumee, Ohio) was used for the next 8 months. Aspen bedding was used for the remainder of the study. Reusable filter bonnets and pipe racks were sanitized every 2 weeks throughout the study.

Water was available from 250 ml polycarbonate water bottles equipped with rubber stoppers and stainless steel sipper tubes. Bottles were replaced twice weekly and, for rats only, water was supplied as needed between changes. Food and water were available _ad libitum_.

Wayne Lab-Blox® meal was supplied to rats for 12 months and mice for 11 months from Alpine® aluminum feed cups (Curtin Matheson Scientific, Inc., Woburn, Massachusetts) containing stainless steel baffles. After that period, meal was supplied from stainless steel gangstyle food hoppers (Scientific Cages, Inc., Bryan, Texas). During the 2-year period of chemical administration, dosed animals were supplied with meal containing the appropriate concentrations of 1,5-naphthalenediamine. Control animals had untreated meal available. Food hoppers were changed on the same schedule as were cages. Food was replenished daily in Alpine® feed cups.

All rats utilized in the 1,5-naphthalenediamine bioassay were housed in a room with other rats receiving diets containing[*] acetylaminofluorene (53-96-3); sodium nitrite (76-32-00-0); L-arginine glutamate (4320-30-3); N-butylurea (592-31-4); N,N-dimethyl-p-nitrosoaniline (138-89-6); 2,5-toluenediamine sulfate (6369-59-1); 2,4-dinitrotoluene (121-14-2); 4-nitroanthranilic acid (619-17-0); N-(1-naphthyl)ethylenediamine dihydrochloride (1465-25-4); 2-chloro-p-phenylenediamine sulfate (61702-44-1); aniline hydrochloride (142-04-1); and p-anisidine hydrochloride (20265-97-8).

Dosed mice were in a room with mice intubated with m-cresidine (102-50-1); and with other mice receiving diets containing N-(1-naphthyl)ethylenediamine dihydrochloride (1465-25-4) and 1H-benzotriazole (95-14-7). Control mice were in a room with other mice receiving diets containing hydrazobenzene (530-50-7); 2,3,5,6-tetrachloro-4-nitroanisole (2438-88-2); tris(2,3-dibromopropyl)phosphate (126-72-7); N-(1-naphthyl)ethylenediamine dihydrochloride (1465-25-4); aniline hydrochloride (142-04-1); and 2-chloro-o-phenylenediamine sulfate.

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[*] CAS registry numbers are given in parentheses.

E. Selection of Initial Concentrations

In order to establish the maximum tolerated concentrations of 1,5-naphthalenediamine for administration to dosed animals in the chronic studies, subchronic toxicity studies were conducted with both rats and mice. Animals of each species were distributed among six groups, each consisting of five males and five females. 1,5-Naphthalenediamine was incorporated into the basal laboratory diet and supplied *ad libitum* to five of the six rat groups and five of the six mouse groups in concentrations of 0.03, 0.1, 0.3, 1.0, and 3.0 percent. The sixth group of each species served as a control group, receiving only the basal laboratory diet. The dosed dietary preparations were administered for 8 weeks.

The highest concentration causing no deaths, no compound-related gross abnormalities, and no mean body weight depression in excess of 20 percent relative to controls was selected as the high concentration for the chronic bioassay.

Deaths were recorded for all groups of rats receiving concentrations of 0.3 percent or more. Mean body weight depression was approximately 19 and 9 percent, respectively, in males and females dosed with 0.1 percent 1,5-naphthalenediamine. The concentration of 1,5-naphthalenediamine selected for administration as the high dose in the rat chronic bioassay was 0.1 percent.

Deaths were recorded for all groups of mice receiving concentrations of 0.3 percent or more and in the group of female mice receiving 0.03 percent. Mean body weight depression was approximately 22 and 3 percent, respectively, in males and females dosed with 0.3 percent. Males receiving 0.1 percent experienced mean body weight depression of approximately 3 percent, while females receiving the same concentration had a greater mean body weight than the controls. The concentration of 1,5-naphthalenediamine selected for administration as the high dose in the mouse chronic bioassay was 0.2 percent.

F. Experimental Design

The experimental design parameters for the chronic study (species, sex, group size, concentrations administered, and duration of treated and untreated observation periods) are summarized in Tables 1 and 2.

Rats were all approximately 7 weeks old at the time they were placed on test. Dosed rats were born approximately 1 month earlier than controls and were started on test 1 month earlier than controls. The dietary concentrations of 1,5-naphthalenediamine administered were 0.10 and 0.05 percent. Throughout this report those rats receiving the former concentration are referred to as the high dose groups and those receiving the latter concentration are referred to as the low dose groups. The dosed rats were supplied with feed containing 1,5-naphthalenediamine for a total of 103 weeks, followed by a 3- to 4-week observation period.

All mice were approximately 7 weeks old at the time they were placed on test. Dosed mice were born approximately 1 month earlier

TABLE 1

DESIGN SUMMARY FOR FISCHER 344 RATS 1,5-NAPHTHALENEDIAMINE FEEDING EXPERIMENT

	INITIAL GROUP SIZE	1,5-NAPHTHALENE-DIAMINE CONCENTRATION (PERCENT)	OBSERVATION PERIOD	
			TREATED (WEEKS)	UNTREATED (WEEKS)
MALE				
CONTROL	25	0	0	109
LOW DOSE	50	0.05	103	
		0		3
HIGH DOSE	50	0.10	103	
		0		3
FEMALE				
CONTROL	25	0	0	110
LOW DOSE	50	0.05	103	
		0		3
HIGH DOSE	50	0.10	103	
		0		4

TABLE 2

DESIGN SUMMARY FOR B6C3F1 MICE
1,5-NAPHTHALENEDIAMINE FEEDING EXPERIMENT

	INITIAL GROUP SIZE	1,5-NAPHTHALENEDIAMINE CONCENTRATION (PERCENT)	OBSERVATION PERIOD	
			TREATED (WEEKS)	UNTREATED (WEEKS)
MALE				
CONTROL	50	0	0	109
LOW DOSE	50	0.1	103	
		0		2
HIGH DOSE	50	0.2	103	
		0		2
FEMALE				
CONTROL	50	0	0	109
LOW DOSE	50	0.1	103	
		0		2
HIGH DOSE	50	0.2	103	
		0		3

than controls and were started on test 1 month earlier than controls. The dietary concentrations of 1,5-naphthalenediamine administered were 0.2 and 0.1 percent. Throughout this report those mice receiving the former concentration are referred to as the high dose groups and those receiving the latter concentration are referred to as the low dose groups. The dosed mice were supplied with feed containing 1,5-naphthalenediamine for a total of 103 weeks, followed by a 2- to 3-week observation period.

G. Clinical and Histopathologic Examinations

Animals were weighed immediately prior to initiation of the experiment. Body weights were recorded twice weekly for the first 12 weeks of the study and at monthly intervals thereafter. From the first day, all animals were inspected twice daily for mortality. Food consumption, for two cages from each group, was monitored for seven consecutive days once a month for the first nine months of the bioassay and for three consecutive days each month thereafter. The presence of tissue masses and lesions was determined by monthly observation and palpation of each animal.

A necropsy was performed on each animal regardless of whether it died, was killed when moribund, or was sacrificed at the end of the bioassay. The animals were euthanized by carbon dioxide inhalation, and were immediately necropsied. The histopathologic examination consisted of gross and microscopic examination of major tissues, organs, and gross lesions taken from sacrificed animals and, whenever possible, from animals found dead.

Tissues were preserved in 10 percent buffered formalin, embedded in paraffin, sectioned, and stained with hematoxylin and eosin prior to microscopic examination. An occasional section was subjected to special staining techniques for more definitive diagnosis.

Slides were prepared from the following tissues: skin, subcutaneous tissue, larynx, lungs and bronchi, trachea, bone marrow, spleen, lymph nodes, thymus, heart, salivary gland, liver, gallbladder (mice), pancreas, esophagus, stomach, small intestine, large intestine, kidney, urinary bladder, pituitary, adrenal, thyroid, parathyroid, ear, brain, testis, prostate, uterus, mammary gland, and ovary.

A few tissues were not examined for some animals, particularly for those that died early. Also, some animals were missing, cannibalized, or judged to be in such an advanced state of autolysis as to preclude histopathologic interpretation. Thus, the number of animals for which particular organs, tissues, or lesions were examined microscopically varies and does not necessarily represent the number of animals that were placed on experiment in each group.

H. Data Recording and Statistical Analyses

Pertinent data on this experiment have been recorded in an automatic data processing system, the Carcinogenesis Bioassay Data System (Linhart et al., 1974). The data elements include descriptive information on the chemicals, animals, experimental design, clinical observations, survival, body weight, and individual pathologic results, as recommended by the International Union Against Cancer (Berenblum, 1969). Data tables were generated for verification of data transcription and for statistical review.

These data were analyzed using the statistical techniques described in this section. Those analyses of the experimental results that bear on the possibility of carcinogenicity are discussed in the statistical narrative sections.

Probabilities of survival were estimated by the product-limit procedure of Kaplan and Meier (1958) and are presented in this report in the form of graphs. Animals were statistically censored as of the time that they died of other than natural causes or were found to be missing; animals dying from natural causes were not statistically censored. Statistical analyses for a possible dose-related effect on survival used the method of Cox (1972) when testing two groups for equality and used Tarone's (1975) extensions of Cox's methods when testing a dose-related trend. One-tailed P-values have been reported for all tests except the departure from linearity test, which is only reported when its two-tailed P-value is less than 0.05.

The incidence of neoplastic or nonneoplastic lesions has been given as the ratio of the number of animals bearing such lesions at a specific anatomic site (numerator) to the number of animals in which that site was examined (denominator). In most instances, the denominators included only those animals for which that site was examined histologically. However, when macroscopic examination was required to detect lesions prior to histologic sampling (e.g., skin or mammary tumors), or when lesions could have appeared at multiple sites (e.g., lymphomas), the denominators consist of the numbers of animals necropsied.

The purpose of the statistical analyses of tumor incidence is to determine whether animals receiving the test chemical developed a significantly higher proportion of tumors than did the control animals. As a part of these analyses, the one-tailed Fisher exact test (Cox, 1970, pp. 48-52) was used to compare the tumor incidence of a control group to that of a group of treated animals at each dose level. When results for a number of treated groups, k, are compared simultaneously with those for a control group, a correction to ensure an overall significance level of 0.05 may be made. The Bonferroni inequality (Miller, 1966, pp. 6-10) requires that the P-value for any comparison be less than or equal to 0.05/k. In cases where this correction was used, it is discussed in the narrative section. It is not, however, presented in the tables, where the Fisher exact P-values are shown.

The Cochran-Armitage test for linear trend in proportions, with continuity correction (Armitage, 1971, pp. 362-365), was also used when appropriate. Under the assumption of a linear trend, this test determined if the slope of the dose-response curve is different from zero at the one-tailed 0.05 level of significance. Unless otherwise noted, the direction of the significant trend was a positive dose relationship. This method also provides a two-tailed test of departure from linear trend.

A time-adjusted analysis was applied when numerous early deaths resulted from causes that were not associated with the formation of tumors. In this analysis, deaths that occurred before the first tumor was observed were excluded by basing the statistical tests on animals that survived at least 52 weeks, unless a tumor was found at the anatomic site of interest before week 52. When such an early tumor was found, comparisons were based exclusively on animals that survived at least as long as the animal in which the first tumor was found. Once this reduced set of data was obtained, the standard procedures for analyses of the incidence of tumors (Fisher exact tests, Cochran-Armitage tests, etc.) were followed.

When appropriate, life-table methods were used to analyze the incidence of tumors. Curves of the proportions surviving without an observed tumor were computed as in Saffiotti et al. (1972). The week during which animals died naturally or were sacrificed was entered as the time point of tumor observation. Cox's methods of comparing these curves were used for two groups; Tarone's extension to testing for linear trend was used for three groups. The statistical tests for the incidence of tumors which used life-table methods were one-tailed and, unless otherwise noted, in the direction of a positive dose relationship. Significant departures from linearity ($P < 0.05$, two-tailed test) were also noted.

The approximate 95 percent confidence interval for the relative risk of each dosed group compared to its control was calculated from the exact interval on the odds ratio (Gart, 1971). The relative risk is defined as p_t/p_c where p_t is the true binomial probability of the incidence of a specific type of tumor in a treated group of animals and p_c is the true probability of the spontaneous incidence of the same type of tumor in a control group. The hypothesis of equality between the true proportion of a specific tumor in a treated group and the proportion in a control group corresponds to a relative risk of unity. Values in excess of unity represent the condition of a larger proportion in the treated group than in the control.

The lower and upper limits of the confidence interval of the relative risk have been included in the tables of statistical analyses. The interpretation of the limits is that in approximately 95 percent of a large number of identical experiments, the true ratio of the risk in a treated group of animals to that in a control group would be within the interval calculated from the experiment. When the lower limit of the confidence interval is greater than one, it can be inferred that a statistically significant result (a $P < 0.025$ one-tailed test when the control incidence is not zero, $P < 0.050$ when the control incidence is zero) has occurred. When the lower limit is less than unity but the upper limit is greater than unity, the lower limit indicates the absence of a significant result while the upper limit indicates that there is a theoretical possibility of the induction of tumors by the test chemical which could not be detected under the conditions of this test.

III. CHRONIC TESTING RESULTS: RATS

A. Body Weights and Clinical Observations

There was no appreciable depression in mean body weight when dosed rats were compared with their respective controls (Figure 2).

Subcutaneous masses were observed in 2 high dose, 3 low dose, and 1 control males, and in 12 high dose, 3 low dose, and 2 control females. Crusted cutaneous masses occurred in 4 high dose males, 1 low dose male, 2 low dose females, and 1 control female, while firm nodular growths were detected in 1 high dose, 2 low dose, and 2 control males, and in 1 low dose female. Swelling of the eyes was exhibited by 2 high dose males, 2 high dose females, and 2 low dose females and swelling of the nose by 1 low dose male. Only 1 control female experienced crusted lesions in the vaginal area while 4 low dose and 9 high dose females were so effected. Alopecia was recorded for 1 low dose female, emaciation was observed in 1 male and 1 female control, and 1 female control exhibited abdominal distention.

B. Survival

The estimated probabilities of survival for male and female rats in the control and 1,5-naphthalenediamine-dosed groups are shown in Figure 3. There was no significant positive association between dosage and mortality for either male or female rats.

Adequate numbers of male rats were at risk from late-developing tumors with 74 percent (37/50) of the high dose, 80 percent (40/50) of the low dose and 68 percent (17/25) of the control surviving on

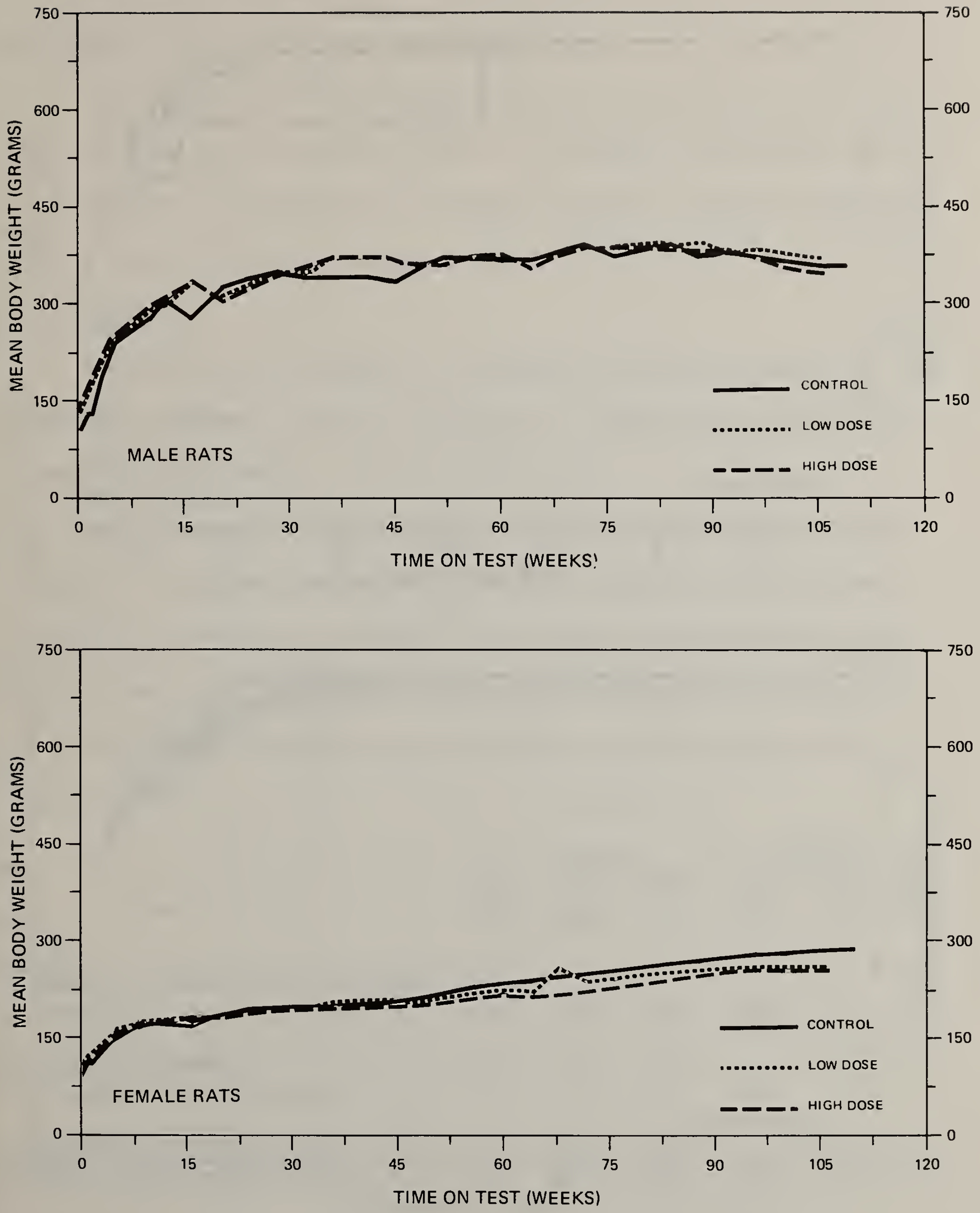
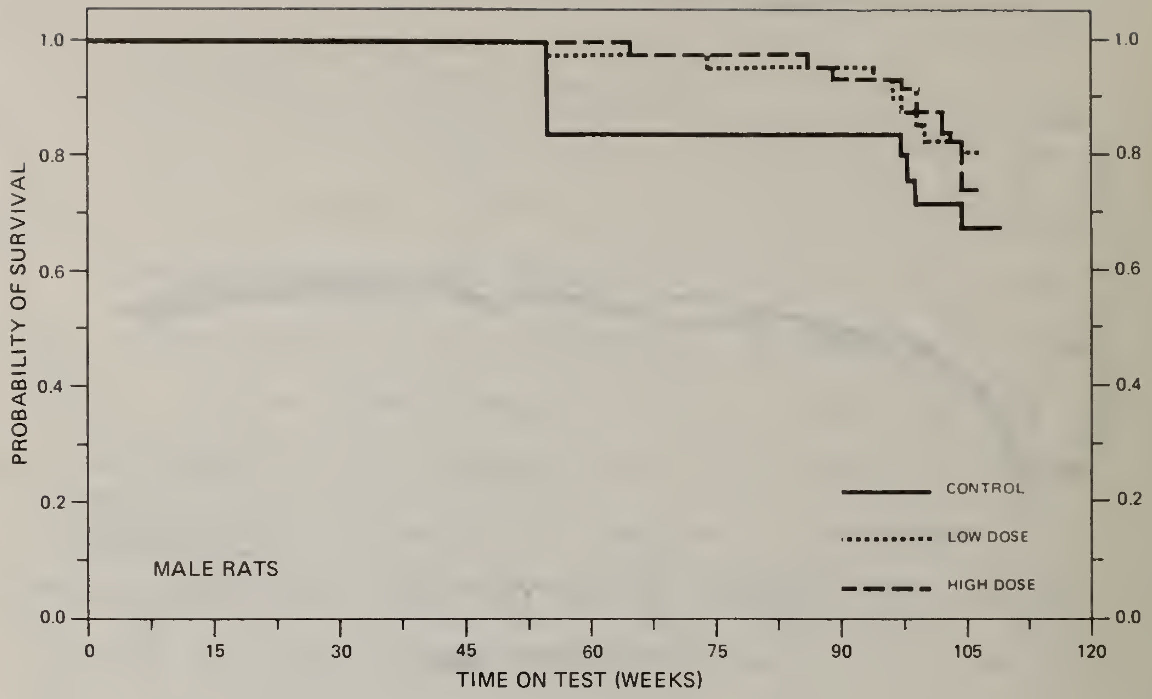


FIGURE 2
GROWTH CURVES FOR 1,5-NAPHTHALENEDIAMINE CHRONIC STUDY RATS



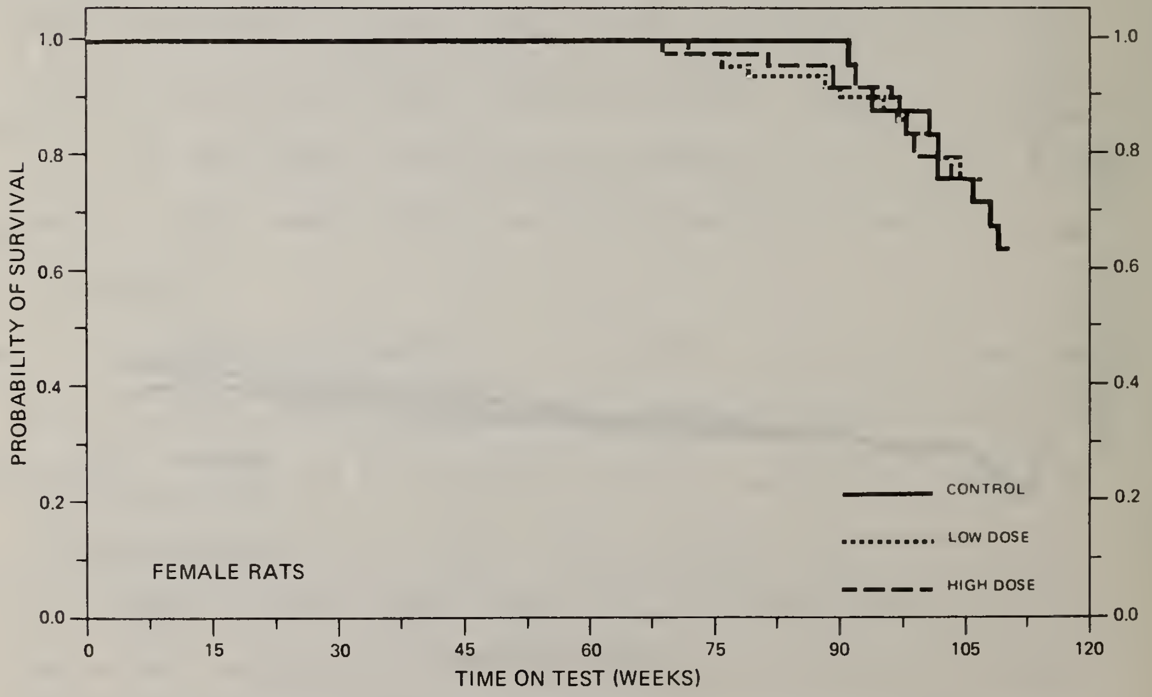


FIGURE 3
SURVIVAL COMPARISONS OF 1,5-NAPHTHALENEDIAMINE CHRONIC STUDY RATS

test until the termination of the study. No lesions were reported for the 4 control rats that died in week 55.

With 76 percent (38/50) of the high dose, 76 percent (38/50) of the low dose and 64 percent (16/25) of the control rats surviving on test until the termination of the study, adequate numbers of females were at risk from late-developing tumors.

C. Pathology

Histopathologic findings on neoplasms in rats are summarized in Appendix A (Tables A1 and A2); findings on nonneoplastic lesions are summarized in Appendix C (Tables C1 and C2).

The incidence of liver neoplasms in male and female rats administered 1,5-naphthalenediamine in the diet appeared to be increased relative to controls. In female rats, tumors of the clitoral gland, uterus, and C-cell neoplasms of the thyroid appeared to be related to compound administration. The incidences of these tumors are as follows:

	MALES			FEMALES		
	Control	Low Dose	High Dose	Control	Low Dose	High Dose
LIVER						
(Number of animals with tissues examined histopathologically)	(25)	(49)	(49)	(24)	(50)	(49)
Neoplastic Nodule	1	3	2	0	3	4
Hepatocellular Carcinoma	0	4	2	0	1	0
PREPUTIAL/CLITORAL GLAND						
(Number of animals necropsied)	(25)	(49)	(50)	(24)	(50)	(50)
Carcinoma	0	0	1	1	3	8
Adenoma	0	0	1	0	0	5

	MALES			FEMALES		
	Control	Low Dose	High Dose	Control	Low Dose	High Dose
UTERUS AND ENDOMETRIUM (Number of animals with tissues examined histopathologically)	-	-	-	(24)	(49)	(48)
Adenocarcinoma				1	2	4
Endometrial Stromal Polyp				2	14	20
Endometrial Stromal Sarcoma				1	2	2
THYROID (Number of animals with tissues examined histopathologically)	(21)	(47)	(47)	(21)	(49)	(48)
C-Cell Adenoma	0	2	5	0	7	3
C-Cell Carcinoma	2	3	3	1	5	1

Neoplasms of the clitoral (preputial) gland were presented grossly as round, fluctuant cystic subcutaneous lesions in the genital area, which on section were filled with pasty green material. On microscopic examination, the cyst contents consisted of desquamated epithelial cells, frequently mixed with leukocytes from secondary inflammation. The inner portion of the cyst wall was lined by hyperkeratinized squamous epithelium often thrown into papillary folds. Peripheral to this was a zone of large, round glandular cells at least a few of which had coarse, brightly eosinophilic cytoplasmic granules. If the peripheral border appeared smooth and intact, the lesion was classified as an adenoma. If there was disorganization of the glandular structure and invasion into the surrounding stroma, the tumor was called a carcinoma.

Thyroid C-cell tumors were observed in dosed female rats at incidences increased relative to controls (4/48 [8 percent] high dose, 12/49 [24 percent] low dose, 1/21 [5 percent] controls). C-cell adenomas were discrete masses of these cells, often containing small cysts lined by flat epithelium and containing colloid-like material. In C-cell carcinomas, the tumor cells often assumed a spindle shape and tended to invade surrounding tissue.

Uterine horns containing neoplasms were usually grossly enlarged. The neoplasms themselves were varicolored, polypoid, frequently gelatinous masses projecting into the uterine cavity. Endometrial stromal polyps had a fibrous connective tissue core richly supplied with large vessels. The surface of the polyps was covered with well-differentiated endometrium which often formed glands in the superficial portion of the polyps. These tumors frequently became necrotic at the tip and exhibited hemorrhage and secondary inflammation. In a few rats, the connective tissue stroma of these lesions underwent malignant transformation characterized by increased cellularity, mitoses, and formation of plump, pleomorphic nuclei. Such tumors were classified as stromal sarcomas. A uterine adenocarcinoma was a collection of fairly well-differentiated glands arranged back-to-back with no obvious intervening stroma. Nuclei of the glands were markedly pleomorphic with frequent mitoses. There was invasion into the myometrium and sometimes into extra uterine structures.

There were instances in this study, as noted in the summary tables, where neoplastic lesions occurred only in dosed animals, or with increased frequency when compared to the control group. No pulmonary neoplasms were found in the controls; alveolar/bronchiolar tumors were seen in dosed rats of both sexes. There was only one urinary tract neoplasm in a female control; a few more occurred in dosed rats, both male and female. No gliomas of the brain were seen in controls; a few gliomas were found in dosed rats of both sexes. These neoplasms occurred in such small numbers that a conclusive interpretation as to their significance is not possible.

Rats in all groups exhibited a variety of nonneoplastic inflammatory and degenerative changes, and none were associated with administration of the compound.

Based upon the results of this pathologic examination, 1,5-naphthalenediamine was carcinogenic to female Fischer 344 rats since feeding of the compound was associated with adenomas and carcinomas of the clitoral gland. In addition, 1,5-naphthalenediamine feeding appeared to be associated with increased incidences of thyroid, liver and uterine neoplasms in female rats and liver neoplasms in male rats.

D. Statistical Analyses of Results

The results of the statistical analyses of tumor incidence in rats are summarized in Tables 3 and 4. The analysis is included for

TABLE 3

ANALYSES OF THE INCIDENCE OF PRIMARY TUMORS AT
SPECIFIC SITES IN MALE RATS TREATED WITH 1,5-NAPHTHALENEDIAMINE[a]

TOPOGRAPHY:MORPHOLOGY	CONTROL	LOW DOSE	HIGH DOSE
Subcutaneous Tissue: Fibroma[b]	1/25(0.04)	3/49(0.06)	2/50(0.04)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	1.531	1.000
Lower Limit	---	0.133	0.056
Upper Limit	---	78.493	56.712
Weeks to First Observed Tumor	99	106	102
Skin: Squamous-Cell Papilloma[b]	2/25(0.08)	1/49(0.02)	1/50(0.02)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	0.255	0.250
Lower Limit	---	0.005	0.004
Upper Limit	---	4.707	4.616
Weeks to First Observed Tumor	109	106	106
Lung: Alveolar/Bronchiolar Adenoma or Alveolar/Bronchiolar Carcinoma[b]	0/25(0.00)	3/49(0.06)	4/47(0.09)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	Infinite	Infinite
Lower Limit	---	0.315	0.508
Upper Limit	---	Infinite	Infinite
Weeks to First Observed Tumor	---	104	106

TABLE 3 (CONTINUED)

TOPOGRAPHY:MORPHOLOGY	CONTROL	LOW DOSE	HIGH DOSE
Hematopoietic System: Leukemia or Malignant Lymphoma[b]	1/25(0.04)	10/49(0.20)	10/50(0.20)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	5.102	5.000
Lower Limit	---	0.801	0.787
Upper Limit	---	212.137	213.351
Weeks to First Observed Tumor	109	100	97
Liver: Hepatocellular Carcinoma or Neoplastic Nodule[b]	1/25(0.04)	7/49(0.14)	4/49(0.08)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	3.571	2.041
Lower Limit	---	0.503	0.218
Upper Limit	---	156.046	96.949
Weeks to First Observed Tumor	109	106	104
Pituitary: Adenoma NOS, Chromophobe Adenoma, Acidophil Adenoma, or Basophil Adenoma[b]	2/22(0.09)	7/44(0.16)	11/44(0.25)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	1.750	2.750
Lower Limit	---	0.376	0.683
Upper Limit	---	16.365	24.081
Weeks to First Observed Tumor	98	96	65

TABLE 3 (CONTINUED)

TOPOGRAPHY:MORPHOLOGY	CONTROL	LOW DOSE	HIGH DOSE
Adrenal: Pheochromocytoma or Malignant Pheochromocytoma[b]	2/24(0.08)	4/48(0.08)	5/48(0.10)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	1.000	1.250
Lower Limit	---	0.157	0.226
Upper Limit	---	10.563	12.529
Weeks to First Observed Tumor	109	106	102
Thyroid: C-Cell Carcinoma[b]	2/21(0.10)	3/47(0.06)	3/47(0.06)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	0.670	0.670
Lower Limit	---	0.084	0.084
Upper Limit	---	7.650	7.650
Weeks to First Observed Tumor	97	100	106
Thyroid: C-Cell Adenoma or C-Cell Carcinoma[b]	2/21(0.10)	5/47(0.11)	8/47(0.17)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	1.117	1.787
Lower Limit	---	0.205	0.405
Upper Limit	---	11.249	16.445
Weeks to First Observed Tumor	97	100	104

TABLE 3 (CONCLUDED)

TOPOGRAPHY:MORPHOLOGY	CONTROL	LOW DOSE	HIGH DOSE
Pancreatic Islets: Islet-Cell Adenoma or Islet-Cell Carcinoma[b]	1/25(0.04)	2/48(0.04)	5/45(0.11)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	1.042	2.778
Lower Limit	---	0.058	0.340
Upper Limit	---	60.184	128.213
Weeks to First Observed Tumor	98	106	104
Testis: Interstitial-Cell Tumor[b]	21/25(0.84)	44/49(0.90)	45/49(0.92)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	1.069	1.093
Lower Limit	---	0.890	0.912
Upper Limit	---	1.325	1.324
Weeks to First Observed Tumor	97	94	65

[a] Treated groups received doses of 0.05 or 0.10 percent in feed.

[b] Number of tumor-bearing animals/number of animals examined at site (proportion).

[c] The probability level for the Cochran-Armitage test is given beneath the incidence of tumors in the control group when $P < 0.05$; otherwise, not significant (N.S.) is indicated. The probability level for the Fisher exact test for the comparison of a treated group with the control group is given beneath the incidence of tumors in the treated group when $P < 0.05$; otherwise, not significant (N.S.) is indicated. For both Cochran-Armitage and Fisher exact tests a negative designation (N) indicates a lower incidence in the treated group(s) than in the control group.

[d] The 95% confidence interval on the relative risk of the treated group to the control group.

TABLE 4

ANALYSES OF THE INCIDENCE OF PRIMARY TUMORS AT SPECIFIC SITES IN FEMALE RATS TREATED WITH 1,5-NAPHTHALENEDIAMINE[a]

TOPOGRAPHY:MORPHOLOGY	CONTROL	LOW DOSE	HIGH DOSE
Hematopoietic System: Leukemia or Malignant Lymphoma[b]	3/24(0.13)	7/50(0.14)	1/50(0.02)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	1.120	0.160
Lower Limit	---	0.287	0.003
Upper Limit	---	6.292	1.890
Weeks to First Observed Tumor	94	76	103
Liver: Hepatocellular Carcinoma or Neoplastic Nodule[b]	0/24(0.00)	4/50(0.08)	4/49(0.08)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	Infinite	Infinite
Lower Limit	---	0.458	0.467
Upper Limit	---	Infinite	Infinite
Weeks to First Observed Tumor	---	102	106
Pituitary: Adenoma NOS, Chromophobe Adenoma, Acidophil Adenoma, or Basophil Adenoma[b]	6/21(0.29)	10/50(0.20)	17/47(0.36)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	0.700	1.266
Lower Limit	---	0.275	0.577
Upper Limit	---	2.090	3.426
Weeks to First Observed Tumor	91	98	98

TABLE 4 (CONTINUED)

TOPOGRAPHY:MORPHOLOGY	CONTROL	LOW DOSE	HIGH DOSE
Pituitary: Carcinoma NOS, Adenoma NOS, Chromophobe Adenoma, Chromophobe Carcinoma, Acidophil Adenoma, or Basophil Adenoma[b]	6/21(0.29)	11/50(0.22)	18/47(0.38)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	0.770	1.340
Lower Limit	---	0.312	0.618
Upper Limit	---	2.262	3.606
Weeks to First Observed Tumor	91	88	98
Adrenal: Cortical Adenoma or Cortical Carcinoma[b]	0/24(0.00)	3/50(0.06)	1/49(0.02)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	Infinite	Infinite
Lower Limit	---	0.297	0.027
Upper Limit	---	Infinite	Infinite
Weeks to First Observed Tumor	---	106	106
Adrenal: Pheochromocytoma[b]	1/24(0.04)	0/50(0.00)	3/49(0.06)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	0.000	1.469
Lower Limit	---	0.000	0.127
Upper Limit	---	8.966	75.534
Weeks to First Observed Tumor	110	---	106

TABLE 4 (CONTINUED)

TOPOGRAPHY:MORPHOLOGY	CONTROL	LOW DOSE	HIGH DOSE
Thyroid: C-Cell Carcinoma[b]	1/21(0.05)	5/49(0.10)	1/48(0.02)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	2.143	0.438
Lower Limit	---	0.266	0.006
Upper Limit	---	99.147	33.659
Weeks to First Observed Tumor	109	106	106
Thyroid: C-Cell Adenoma or C-Cell Carcinoma[b]	1/21(0.05)	12/49(0.24)	4/48(0.08)
P Values[c]	N.S.	P = 0.046	N.S.
Departure from Linear Trend[e]	P = 0.009	---	---
Relative Risk (Control)[d]	---	5.143	1.750
Lower Limit	---	0.855	0.192
Upper Limit	---	215.370	83.548
Weeks to First Observed Tumor	109	104	103
Thyroid: Papillary Carcinoma, Follicular-Cell Carcinoma, or Papillary Cystadenocarcinoma NOS[b]	1/21(0.05)	1/49(0.02)	3/48(0.06)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	0.429	1.313
Lower Limit	---	0.006	0.115
Upper Limit	---	32.983	67.452
Weeks to First Observed Tumor	110	106	99

TABLE 4 (CONTINUED)

TOPOGRAPHY:MORPHOLOGY	CONTROL	LOW DOSE	HIGH DOSE
Thyroid: Papillary Carcinoma, Follicular-Cell Carcinoma, Papillary Cystadenocarcinoma NOS, or Papillary Cystadenoma[b]	1/21(0.05)	2/49(0.04)	4/48(0.08)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	0.857	1.750
Lower Limit	---	0.648	0.191
Upper Limit	---	49.555	84.310
Weeks to First Observed Tumor	110	106	81
Mammary Gland: Fibroadenoma[b]	4/24(0.17)	5/50(0.10)	13/50(0.26)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	0.600	1.560
Lower Limit	---	0.145	0.556
Upper Limit	---	2.812	6.019
Weeks to First Observed Tumor	109	102	98
Mammary Gland: Fibroadenoma, Adenocarcinoma NOS, or Papillary Adenocarcinoma[b]	4/24(0.17)	5/50(0.10)	14/50(0.28)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	0.600	1.680
Lower Limit	---	0.145	0.609
Upper Limit	---	2.807	6.412
Weeks to First Observed Tumor	109	102	98

TABLE 4 (CONTINUED)

TOPOGRAPHY:MORPHOLOGY	CONTROL	LOW DOSE	HIGH DOSE
Clitoral Gland: Carcinoma NOS[b]	1/24(0.04)	3/50(0.06)	8/50(0.16)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	1.440	3.840
Lower Limit	---	0.125	0.566
Upper Limit	---	75.487	168.221
Weeks to First Observed Tumor	110	106	69
Clitoral Gland: Adenoma NOS or Carcinoma NOS[b]	1/24(0.04)	3/50(0.06)	13/50(0.26)
P Values[c]	P = 0.003	N.S.	P = 0.021
Relative Risk (Control)[d]	---	1.440	6.240
Lower Limit	---	0.125	1.043
Upper Limit	---	74.077	258.268
Weeks to First Observed Tumor	110	106	69
Uterus: Endometrial Stromal Polyp[b]	2/24(0.08)	14/49(0.29)	20/48(0.42)
P Values[c]	P = 0.003	P = 0.043	P = 0.003
Relative Risk (Control)[d]	---	3.429	5.000
Lower Limit	---	0.892	1.385
Upper Limit	---	29.588	41.202
Weeks to First Observed Tumor	102	88	96

TABLE 4 (CONCLUDED)

TOPOGRAPHY:MORPHOLOGY	CONTROL	LOW DOSE	HIGH DOSE
Uterus and Endometrium: Adenocarcinoma NOS[b]	1/24(0.04)	2/49(0.04)	4/48(0.08)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	0.980	2.000
Lower Limit	---	0.054	0.216
Upper Limit	---	56.627	96.367
Weeks to First Observed Tumor	110	104	106
Zymbal's Gland: Sebaceous Adenocarcinoma[b]	0/24(0.00)	0/50(0.00)	3/50(0.06)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	---	Infinite
Lower Limit	---	---	0.296
Upper Limit	---	---	Infinite
Weeks to First Observed Tumor	---	---	89

[a] Treated groups received doses of 0.05 or 0.10 percent in feed.

[b] Number of tumor-bearing animals/number of animals examined at site (proportion).

[c] The probability level for the Cochran-Armitage test is given beneath the incidence of tumors in the control group when $P < 0.05$; otherwise, not significant (N.S.) is indicated. The probability level for the Fisher exact test for the comparison of a treated group with the control group is given beneath the incidence of tumors in the treated group when $P < 0.05$; otherwise, not significant (N.S.) is indicated. For both Cochran-Armitage and Fisher exact tests a negative designation (N) indicates a lower incidence in the treated group(s) than in the control group.

[d] The 95% confidence interval on the relative risk of the treated group to the control group.

[e] The probability level of the test for departure from linear trend is given beneath the control group when $P < 0.05$.

every type of tumor in either sex where at least two such tumors were observed in at least one of the control or 1,5-naphthalenediamine-dosed groups and where such tumors were observed in at least 5 percent of the group.

For female rats an increased incidence of endometrial stromal polyps was observed in both the high and low dose groups compared to the control group. The Cochran-Armitage test indicated a significant ($P = 0.003$) positive association between compound administration and tumor incidence. The Fisher exact tests supported this result with a significant ($P = 0.003$) comparison of the high dose group to the control; for the low dose comparison the probability level was $P = 0.043$, a marginal result which was not significant under the Bonferroni criterion. Based on these results, the administration of 1,5-naphthalenediamine was associated with an elevated incidence of endometrial stromal polyps in female rats.

A number of adenomas NOS and carcinomas NOS of the clitoral gland were observed in female rats. The Cochran-Armitage test indicated a significant ($P = 0.003$) positive association between dose and the combined incidence of adenomas NOS or carcinomas NOS of the clitoral gland. The Fisher exact test comparing high dose to control was also significant ($P = 0.021$). In historical data collected by this laboratory for the NCI Carcinogenesis Testing Program, 4/249 (2 percent) of the untreated female Fischer 344 rats had one of these tumors, compared to the 13/50 (26 percent) observed in the high dose group in this bioassay. Based upon these statistical results, the administration of 1,5-naphthalenediamine was associated with an elevated incidence of clitoral gland neoplasms in female rats.

For females the Fisher exact test comparing control to low dose for the combined incidence of C-cell adenomas or C-cell carcinomas of the thyroid had a probability level of P = 0.046, a marginal result which was not significant under the Bonferroni criterion.

Based on these statistical tests, it is concluded that 1,5-naphthalenediamine was carcinogenic for female rats, producing tumors of the clitoral gland and uterus.

IV. CHRONIC TESTING RESULTS: MICE

A. Body Weights and Clinical Observations

Mean body weight depression was readily apparent in dosed male mice when compared to controls. A similar but less pronounced trend was evident in dosed females (Figure 4).

One low dose male had a soft subcutaneous mass on the leg and two males in this group had palpable abdominal masses. Firm nodular growths developed in one low dose male and two high dose females. Alopecia was observed in 27 control males, 16 low dose males, 4 high dose males, 25 control females, and 3 low dose females. Two low dose and two high dose males experienced noticeable swelling of the eyes. Abdominal distention was observed in one control male and one control female mouse.

B. Survival

The estimated probabilities of survival for male and female mice in the control and 1,5-naphthalenediamine-dosed groups are shown in Figure 5. There was no significant positive association between dosage and mortality for either male or female mice.

Adequate numbers of male mice were at risk from late-developing tumors with 58 percent (29/50) of the high dose, 78 percent (39/50) of the low dose and 66 percent (33/50) of the controls surviving on test until the termination of the study. The 6 control male mice that died in week 11 were autolyzed, as were 2 of the 4 high dose male mice that died in week 41.

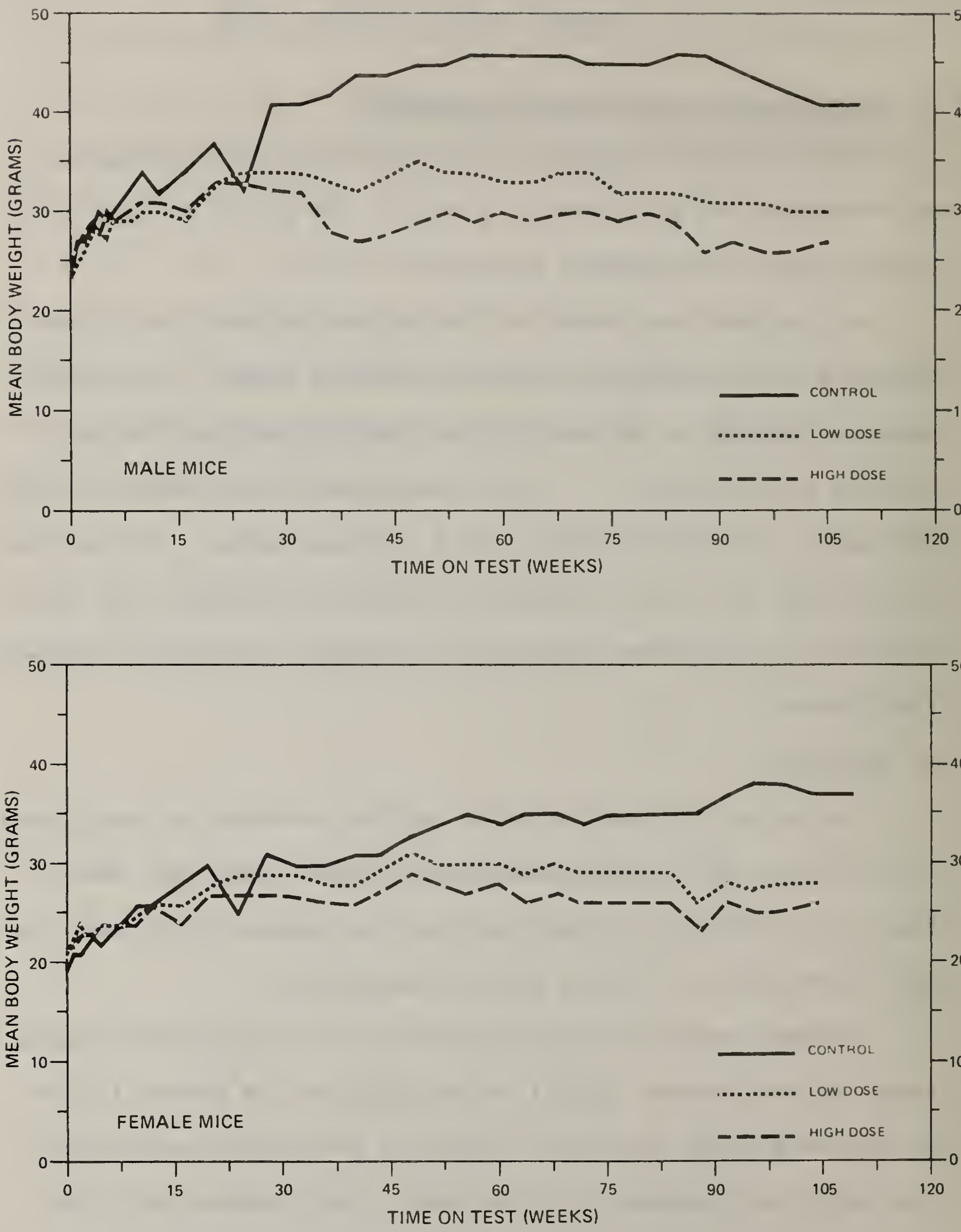
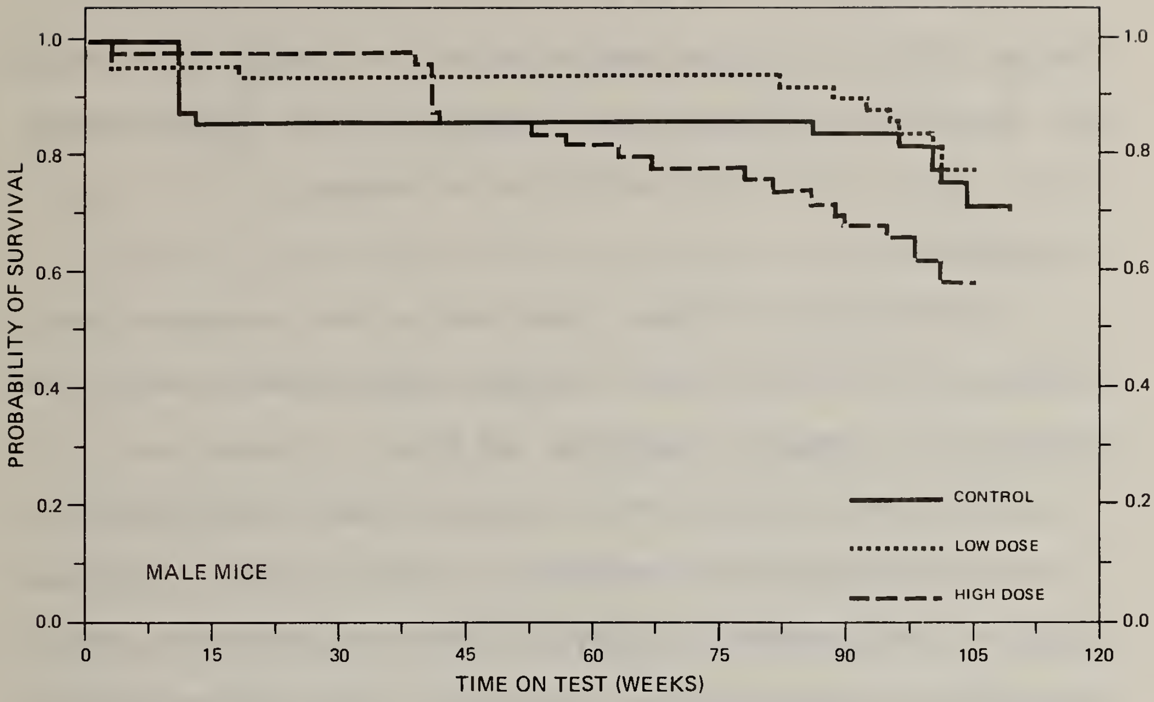


FIGURE 4
GROWTH CURVES FOR 1,5-NAPHTHALENEDIAMINE CHRONIC STUDY MICE



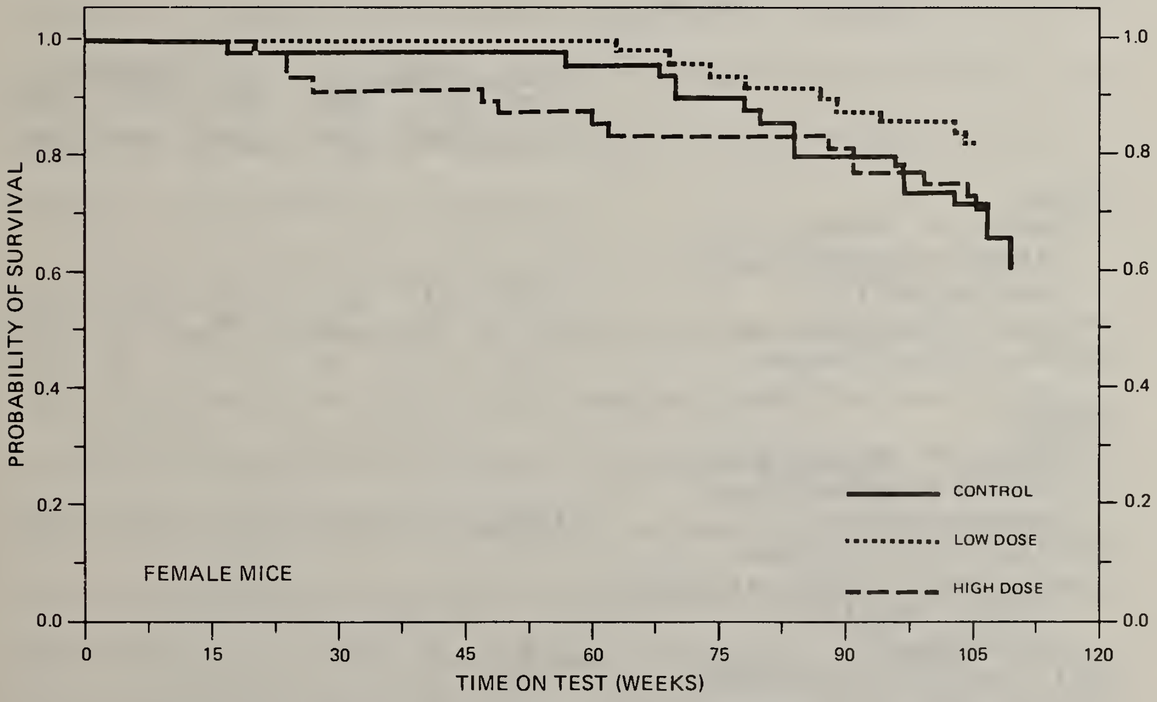


FIGURE 5
SURVIVAL COMPARISONS OF 1,5-NAPHTHALENEDIAMINE CHRONIC STUDY MICE

For female mice, with 68 percent (34/50) of the high dose, 82 percent (41/50) of the low dose and 60 percent (30/50) of the control mice surviving on test until the termination of the study, adequate numbers were at risk from late-developing tumors.

C. Pathology

Histopathologic findings on neoplasms in mice are summarized in Appendix B (Tables B1 and B2); findings on nonneoplastic lesions are summarized in Appendix D (Tables D1 and D2).

Dietary administration of 1,5-naphthalenediamine produced an increase in hepatocellular neoplasms in female mice, and it produced a dose-related increase in thyroid neoplasms and compound-related nonneoplastic thyroid lesions in both sexes. The compound-related lesions are summarized below:

	MALES			FEMALES		
	Control	Low Dose	High Dose	Control	Low Dose	High Dose
LIVER						
(Number of animals with tissues examined histopathologically)	(39)	(45)	(43)	(46)	(49)	(46)
Hepatocellular Carcinoma	12	10	7	1	25	16
Hepatocellular Adenoma	0	3	6	0	3	11
THYROID						
(Number of animals with tissues examined histopathologically)	(38)	(46)	(43)	(44)	(49)	(45)
Follicular-Cell Adenoma (Papillary or Follicular-Cell Adenoma, Papillary Cystadenoma)	0	8	16	2	17	14
Follicular-Cell Carcinoma	0	1	1	2	0	1
Follicular-Cell Hyperplasia	2	12	9	2	1	4
C-Cell Adenoma	0	2	0	0	1	2
C-Cell Carcinoma	0	0	4	0	1	6

In male mice, dietary administration of the compound did not increase the incidence of hepatocellular neoplasms, whereas dosed females showed a striking increase in hepatocellular carcinomas and hepatocellular adenomas.

Grossly, hepatocellular neoplasms appeared as smooth, nodular, rounded masses distorting the normal shape of the liver. Color varied, many neoplasms appearing pale tan or dark red. Microscopically, hepatocellular carcinomas were expansive masses of hepatocytes exhibiting loss of normal architectural pattern, the cells being arranged in sheets or trabeculae instead of the normal lobules. Nuclei were frequently uniform, although variable amounts of pleomorphism did occur. The cytoplasm was either basophilic or acidophilic, sometimes varying from one region of the tumor to another, and was frequently pale. Lesions classified as hepatocellular adenomas were smaller, usually better differentiated, and were less pleomorphic than the hepatocellular carcinomas.

The criteria for classification of thyroid neoplasms in mice were the same as those used to classify thyroid neoplasms in rats. The nonneoplastic thyroid lesions found in dosed mice were similar to those in the rats but occurred in higher incidences. Hyperplasia of follicular cells (focal, papillary or adenomatous) were found in 2/38 (5 percent) control, 12/46 (26 percent) low dose, and 9/43 (21 percent) high dose male mice. Abundant golden brown pigment was seen in follicular epithelium, colloid, and macrophages. In the mice, there were frequent foci of lymphocytes in the thyroid parenchyma and occasional cystic areas filled with amorphous material containing long clefts suggesting cholesterol crystals.

Three transitional-cell papillomas occurred in the bladder or urethra of dosed mice (two high dose males and one high dose female), but none occurred in controls.

Based upon the results of this pathologic examination, 1,5-naphthalenediamine was carcinogenic to B6C3F1 mice, producing hepatocellular neoplasms in females and thyroid neoplasms in both sexes.

D. Statistical Analyses of Results

The results of the statistical analyses of tumor incidence in mice are summarized in Tables 5 and 6. The analysis is included for every type of tumor in either sex where at least two such tumors were observed in at least one of the control or 1,5-naphthalenediamine-dosed groups and where such tumors were observed in at least 5 percent of the group.

For both male and female mice elevated incidences of thyroid tumors were observed in the dosed groups. In female mice the Cochran-Armitage test indicated a significant ($P = 0.005$) positive association between dietary concentration and the incidence of C-cell carcinomas. This was supported by a significant ($P = 0.014$) Fisher exact test for the high dose group. For males the Cochran-Armitage test result was also significant ($P = 0.017$), but the Fisher exact tests were

TABLE 5

ANALYSES OF THE INCIDENCE OF PRIMARY TUMORS AT SPECIFIC SITES IN MALE MICE TREATED WITH 1,5-NAPHTHALENEDIAMINE[a]

TOPOGRAPHY:MORPHOLOGY	CONTROL	LOW DOSE	HIGH DOSE
Lung: Alveolar/Bronchiolar Carcinoma[b]	2/39(0.05)	3/46(0.07)	0/45(0.00)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	1.272	0.000
Lower Limit	---	0.153	0.000
Upper Limit	---	14.686	4.478
Weeks to First Observed Tumor	109	82	---
Lung: Alveolar/Bronchiolar Adenoma or Alveolar/Bronchiolar Carcinoma[b]	4/39(0.10)	9/46(0.20)	2/45(0.04)
P Values[c]	N.S.	N.S.	N.S.
Departure from Linear Trend[e]	P = 0.037	---	---
Relative Risk (Control)[d]	---	1.908	0.433
Lower Limit	---	0.582	0.041
Upper Limit	---	7.882	2.871
Weeks to First Observed Tumor	109	82	105
Hematopoietic System: Malignant Lymphoma[b]	13/39(0.33)	14/47(0.30)	5/49(0.10)
P Values[c]	P = 0.007(N)	N.S.	P = 0.008(N)
Relative Risk (Control)[d]	---	0.894	0.306
Lower Limit	---	0.448	0.094
Upper Limit	---	1.817	0.829
Weeks to First Observed Tumor	100	82	95

TABLE 5 (CONTINUED)

TOPOGRAPHY:MORPHOLOGY	CONTROL	LOW DOSE	HIGH DOSE
Liver: Hepatocellular Carcinoma[b]	12/39(0.31)	10/45(0.22)	7/43(0.16)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	0.722	0.529
Lower Limit	---	0.318	0.198
Upper Limit	---	1.620	1.306
Weeks to First Observed Tumor	86	88	105
Liver: Hepatocellular Carcinoma or Hepatocellular Adenoma[b]	12/39(0.31)	13/45(0.29)	13/43(0.30)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	0.939	0.983
Lower Limit	---	0.453	0.473
Upper Limit	---	1.981	2.071
Weeks to First Observed Tumor	86	88	105
Thyroid: C-Cell Carcinoma[b]	0/38(0.00)	0/46(0.00)	4/43(0.09)
P Values[c]	P = 0.017	N.S.	N.S.
Relative Risk (Control)[d]	---	---	Infinite
Lower Limit	---	---	0.825
Upper Limit	---	---	Infinite
Weeks to First Observed Tumor	---	---	105

TABLE 5 (CONCLUDED)

TOPOGRAPHY:MORPHOLOGY	CONTROL	LOW DOSE	HIGH DOSE
Thyroid: C-Cell Carcinoma or C-Cell Adenoma[b]	0/38(0.00)	2/46(0.04)	4/43(0.09)
P Values[c]	P = 0.044	N.S.	N.S.
Relative Risk (Control)[d]	---	Infinite	Infinite
Lower Limit	---	0.246	0.825
Upper Limit	---	Infinite	Infinite
Weeks to First Observed Tumor	---	105	105
Thyroid: Papillary Adenoma, Follicular-Cell Adenoma, or Papillary Cystadenoma NOS[b]	0/38(0.00)	8/46(0.17)	16/43(0.37)
P Values[c]	P < 0.001	P = 0.006	P < 0.001
Relative Risk (Control)[d]	---	Infinite	Infinite
Lower Limit	---	1.905	4.523
Upper Limit	---	Infinite	Infinite
Weeks to First Observed Tumor	---	105	98

[a] Treated groups received doses of 0.1 or 0.2 percent in feed.

[b] Number of tumor-bearing animals/number of animals examined at site (proportion).

[c] The probability level for the Cochran-Armitage test is given beneath the incidence of tumors in the control group when $P < 0.05$; otherwise, not significant (N.S.) is indicated. The probability level for the Fisher exact test for the comparison of a treated group with the control group is given beneath the incidence of tumors in the treated group when $P < 0.05$; otherwise, not significant (N.S.) is indicated. For both Cochran-Armitage and Fisher exact tests a negative designation (N) indicates a lower incidence in the treated group(s) than in the control group.

[e] The probability level of the test for departure from linear trend is given beneath the control group when $P < 0.05$.

TABLE 6

ANALYSES OF THE INCIDENCE OF PRIMARY TUMORS AT SPECIFIC SITES IN FEMALE MICE TREATED WITH 1,5-NAPHTHALENEDIAMINE[a]

TOPOGRAPHY:MORPHOLOGY	CONTROL	LOW DOSE	HIGH DOSE
Lung: Alveolar/Bronchiolar Carcinoma[b]	0/49(0.00)	1/48(0.02)	3/46(0.07)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	Infinite	Infinite
Lower Limit	---	0.055	0.638
Upper Limit	---	Infinite	Infinite
Weeks to First Observed Tumor	---	89	91
Lung: Alveolar/Bronchiolar Adenoma or Alveolar/Bronchiolar Carcinoma[b]	0/49(0.00)	10/48(0.21)	5/46(0.11)
P Values[c]	N.S.	P = 0.001	P = 0.024
Departure from Linear Trend[e]	P = 0.005	---	---
Relative Risk (Control)[d]	---	Infinite	Infinite
Lower Limit	---	3.037	1.347
Upper Limit	---	Infinite	Infinite
Weeks to First Observed Tumor	---	89	91
Hematopoietic System: Leukemia or Malignant Lymphoma[b]	13/49(0.27)	19/50(0.38)	5/46(0.11)
P Values[c]	N.S.	N.S.	P = 0.045(N)
Departure from Linear Trend[e]	P = 0.011	---	---
Relative Risk (Control)[d]	---	1.432	0.410
Lower Limit	---	0.760	0.124
Upper Limit	---	2.781	1.117
Weeks to First Observed Tumor	57	63	105

TABLE 6 (CONTINUED)

TOPOGRAPHY:MORPHOLOGY	CONTROL	LOW DOSE	HIGH DOSE
Liver: Hepatocellular Carcinoma[b]	1/46(0.02)	25/49(0.51)	16/46(0.35)
P Values[c]	$P = 0.001$	$P < 0.001$	$P < 0.001$
Departure from Linear Trend[e]	$P < 0.001$	---	---
Relative Risk (Control)[d]	---	23.469	16.000
Lower Limit	---	4.156	2.683
Upper Limit	---	906.346	646.516
Weeks to First Observed Tumor	109	74	99
Liver: Hepatocellular Adenoma or Hepatocellular Carcinoma[b]	1/46(0.02)	28/49(0.57)	27/46(0.59)
P Values[c]	$P < 0.001$	$P < 0.001$	$P < 0.001$
Departure from Linear Trend[e]	$P = 0.002$	---	---
Relative Risk (Control)[d]	---	26.286	27.000
Lower Limit	---	4.741	4.874
Upper Limit	---	1030.801	1027.943
Weeks to First Observed Tumor	109	74	99
Stomach: Squamous-Cell Papilloma[b]	0/41(0.00)	3/47(0.06)	0/46(0.00)
P Values[c]	N.S.	N.S.	N.S.
Departure from Linear Trend[e]	$P = 0.017$	---	---
Relative Risk (Control)[d]	---	Infinite	---
Lower Limit	---	0.529	---
Upper Limit	---	Infinite	---
Weeks to First Observed Tumor	---	105	---

TABLE 6 (CONTINUED)

TOPOGRAPHY:MORPHOLOGY	CONTROL	LOW DOSE	HIGH DOSE
Pituitary: Adenoma NOS, Chromophobe Adenoma or Acidophil Adenoma[b]	3/34(0.09)	4/35(0.11)	1/30(0.03)
P Values[c]	N.S.	N.S.	N.S.
Relative Risk (Control)[d]	---	1.295	0.378
Lower Limit	---	0.238	0.007
Upper Limit	---	8.188	4.424
Weeks to First Observed Tumor	109	105	106
Adrenal: Pheochromocytoma[b]	3/46(0.07)	0/44(0.00)	0/44(0.00)
P Values[c]	P = 0.040(N)	N.S.	N.S.
Relative Risk (Control)[d]	---	0.000	0.000
Lower Limit	---	0.000	0.000
Upper Limit	---	1.731	1.731
Weeks to First Observed Tumor	68	---	---
Thyroid: C-Cell Carcinoma[b]	0/44(0.00)	1/49(0.02)	6/45(0.13)
P Values[c]	P = 0.005	N.S.	P = 0.014
Relative Risk (Control)[d]	---	Infinite	Infinite
Lower Limit	---	0.048	1.574
Upper Limit	---	Infinite	Infinite
Weeks to First Observed Tumor	---	105	105

TABLE 6 (CONCLUDED)

TOPOGRAPHY:MORPHOLOGY	CONTROL	LOW DOSE	HIGH DOSE
Thyroid: C-Cell Adenoma or C-Cell Carcinoma[b]	0/44(0.00)	2/49(0.04)	8/45(0.18)
P Values[c]	P = 0.001	N.S.	P = 0.003
Relative Risk (Control)[d]	---	Infinite	Infinite
Lower Limit	---	0.267	2.250
Upper Limit	---	Infinite	Infinite
Weeks to First Observed Tumor	---	105	41
Thyroid: Papillary Adenoma, Follicular-Cell Adenoma, or Papillary Cystadenoma NOS[b]	2/44(0.05)	17/49(0.35)	14/45(0.31)
P Values[c]	P = 0.003	P < 0.001	P = 0.001
Departure from Linear Trend[e]	P = 0.025	---	---
Relative Risk (Control)[d]	---	7.633	6.844
Lower Limit	---	1.971	1.709
Upper Limit	---	64.662	58.827
Weeks to First Observed Tumor	80	105	91

[a] Treated groups received doses of 0.1 or 0.2 percent in feed.

[b] Number of tumor-bearing animals/number of animals examined at site (proportion).

[c] The probability level for the Cochran-Armitage test is given beneath the incidence of tumors in the control group when $P < 0.05$; otherwise, not significant (N.S.) is indicated. The probability level for the Fisher exact test for the comparison of a treated group with the control group is given beneath the incidence of tumors in the treated group when $P < 0.05$; otherwise, not significant (N.S.) is indicated. For both Cochran-Armitage and Fisher exact tests a negative designation (N) indicates a lower incidence in the treated group(s) than in the control group.

[d] The 95% confidence interval on the relative risk of the treated group to the control group.

[e] The probability level of the test for departure from linear trend is given beneath the control group when $P < 0.05$.

not. When incidences were combined so that the numerator represented mice with either a papillary adenoma, a follicular-cell adenoma, or a papillary cystadenoma of the thyroid, the Cochran-Armitage test indicated a significant positive association between dietary concentration and tumor incidence for both males ($P < 0.001$) and females ($P = 0.003$). These were supported by significant ($P \leq 0.006$) Fisher exact test results in each sex for comparisons of each dosed group to the control group. Based on these results, the administration of 1,5-naphthalenediamine was associated with the incidence of thyroid neoplasms in both male and female mice.

For females an increased incidence of hepatocellular carcinomas was also observed among the dosed mice. The Cochran-Armitage test indicated a significant ($P = 0.001$) positive association between dose and incidence. This was supported by significant ($P < 0.001$) comparisons of both the high and low dose to the control group using the Fisher exact test. Based on these results the administration of 1,5-naphthalenediamine was associated with the incidence of hepatocellular carcinomas in female mice.

For female mice, when the incidence of alveolar/bronchiolar adenomas and alveolar/bronchiolar carcinomas were combined, an increased incidence in the dosed groups was noted. The Fisher exact test was significant for both the high ($P = 0.024$) and low ($P = 0.001$) dose groups. The departure from linear trend was significant since tumor incidence was increased more in the low dose than in the high dose group. In historical control data compiled by this laboratory for the NCI Carcinogenesis Testing Program, 17/275 (6 percent) of the untreated female B6C3F1 mice had an alveolar/bronchiolar neoplasm. Based upon these results the administration of 1,5-naphthalenediamine was associated with the incidence of alveolar/bronchiolar neoplasms in female mice.

For females the Fisher exact test comparing the incidence of leukemia or malignant lymphoma in high dose mice with that in the controls had a probability level in the negative direction of $P = 0.045$, a marginal result which was not significant under the Bonferroni criterion.

Also for females the Cochran-Armitage test showed a significant ($P = 0.040$) negative association between dose and the incidence of adrenal pheochromocytomas, but the Fisher exact tests were not significant.

In male mice the possibility of a negative association between dose and the incidence of malignant lymphomas or leukemia was noted.

Based upon these statistical results the administration of 1,5-naphthalenediamine was associated with the increased incidence of thyroid neoplasms in male mice and of thyroid neoplasms, of hepatocellular carcinomas, and of alveolar/bronchiolar neoplasms in female mice.

V. DISCUSSION

There were no significant positive associations between dietary concentrations of 1,5-naphthalenediamine and mortality in either sex of rats or mice. In all groups adequate numbers of animals survived sufficiently long to be at risk from late-developing tumors.

Several uterine neoplasms occurred in dosed female rats at higher incidences than in corresponding controls. There was a significant positive association between dietary concentration of the compound and the incidences of endometrial stromal polyps in female rats. In addition, the high dose to control Fisher exact comparison was significant. Endometrial stromal sarcomas were observed in two low dose and two high dose female rats, but not in controls. Uterine adenocarcinomas occurred at a higher incidence in the high dose female rat group than in the control group, but the difference in tumor incidence was not statistically significant.

The administration of 1,5-naphthalenediamine was associated with an elevated incidence of clitoral gland neoplasms in female rats. There was a significant positive association between the concentration of the chemical added to the diet and the incidence of either adenomas or carcinomas of the clitoral gland in female rats. The incidence of either of these neoplasms in the high dose female rat group was significant relative to the incidence in the control group.

Elevated incidences of thyroid neoplasms were observed among dosed mice. For mice of both sexes there were significant positive associations between dietary concentration of 1,5-naphthalenediamine and the incidences of thyroid C-cell carcinomas. For the females the high dose to control Fisher exact comparison supported the finding; this was not true for males. When the mice were grouped so that the numerator of the incidence represented those animals with a papillary adenoma, a follicular-cell adenoma, or a papillary cystadenoma of the thyroid, the Cochran-Armitage test was significantly positive for both males and females and all the Fisher exact comparisons supported the findings.

The incidence of hepatocellular carcinomas in female mice was significantly associated with increased concentration of 1,5-naphthalenediamine. In addition, the high dose to control and the low dose to control Fisher exact comparisons were significant. The incidence of alveolar/bronchiolar adenomas was significant, relative to controls, in both the low dose and the high dose female mouse groups.

Under the conditions of this bioassay, 1,5-naphthalenediamine was carcinogenic in female Fischer 344 rats, causing clitoral and uterine neoplasms. 1,5-Naphthalenediamine was also carcinogenic for B6C3F1 mice, producing thyroid neoplasms in males and neoplasms of the thyroid, liver, and lung in females. Insufficient evidence was provided for the carcinogenicity of the compound in male Fischer 344 rats.

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APPENDIX A

SUMMARY OF THE INCIDENCE OF NEOPLASMS IN RATS TREATED WITH 1,5-NAPHTHALENEDIAMINE

TABLE AI
SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MALE RATS TREATED WITH 1,5-NAPHTHALENEDIAMINE

	CONTROL (UNTR) 01-0330	LOW DOSE 01-0280	HIGH DOSE 01-0285
ANIMALS INITIALLY IN STUDY	25	50	50
ANIMALS NECROPSIED	25	49	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY**	25	49	49
INTEGUMENTARY SYSTEM			
*SKIN	(25)	(49)	(50)
SQUAMOUS CELL PAPILLOMA	2 (8%)	1 (2%)	1 (2%)
SEBACEOUS ADENOCARCINOMA		1 (2%)	
FIBROUS HISTIOCYTOMA		1 (2%)	
*SUBCUT TISSUE	(25)	(49)	(50)
FIBROMA	1 (4%)	3 (6%)	2 (4%)
LIPOMA			1 (2%)
RESPIRATORY SYSTEM			
*LARYNX	(25)	(49)	(50)
PAPILLOMA, NOS			1 (2%)
#TRACHEA	(24)	(16)	(13)
PAPILLOMA, NOS			1 (8%)
#LUNG	(25)	(49)	(47)
ADENOCARCINOMA, NOS, METASTATIC			1 (2%)
ALVEOLAR/BRONCHIOLAR ADENOMA		1 (2%)	4 (9%)
ALVEOLAR/BRONCHIOLAR CARCINOMA		2 (4%)	
C-CELL CARCINOMA, METASTATIC			1 (2%)
SEBACEOUS ADENOCARCINOMA, METAST		1 (2%)	
PHEOCHROMOCYTOMA, METASTATIC	1 (4%)		
HEMATOPOIETIC SYSTEM			
*MULTIPLE ORGANS	(25)	(49)	(50)
LEUKEMIA,NOS			1 (2%)
UNDIFFERENTIATED LEUKEMIA		10 (20%)	4 (8%)
MYELOMONOCYTIC LEUKEMIA	1 (4%)		1 (2%)
LYMPHOCYTIC LEUKEMIA			2 (4%)

# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE A1 (CONTINUED)

	CONTROL(UNTR) 01-0330	LOW DOSE 01-0280	HIGH DOSE 01-0285
#MANDIBULAR L. NODE	(24)	(47)	(47)
C-CELL CARCINOMA, METASTATIC	1 (4%)		
SEBACEOUS ADENOCARCINOMA, METAST		1 (2%)	
NEURILEMOMA, METASTATIC			1 (2%)
#MEDIASTINAL L.NODE	(24)	(47)	(47)
SEBACEOUS ADENOCARCINOMA, METAST		1 (2%)	
#MESENTERIC L. NODE	(24)	(47)	(47)
LYMPHANGIOMA			1 (2%)
#LIVER	(25)	(49)	(49)
UNDIFFERENTIATED LEUKEMIA			1 (2%)
#THYMUS	(13)	(35)	(36)
THYMOMA			1 (3%)
MALIG.LYMPHOMA, LYMPHOCYTIC TYPE			1 (3%)
CIRCULATORY SYSTEM			
NONE			
DIGESTIVE SYSTEM			
#SALIVARY GLAND	(25)	(47)	(46)
ADENOCARCINOMA, NOS			1 (2%)
FIBROSARCOMA		1 (2%)	
NEURILEMOMA, MALIGNANT			1 (2%)
#LIVER	(25)	(49)	(49)
NEOPLASTIC NODULE	1 (4%)	3 (6%)	2 (4%)
HEPATOCELLULAR CARCINOMA		4 (8%)	2 (4%)
LYMPHANGIOMA			1 (2%)
#STOMACH	(24)	(47)	(47)
SQUAMOUS CELL PAPILLOMA		1 (2%)	
URINARY SYSTEM			
#KIDNEY	(25)	(49)	(48)
LIPOMA		1 (2%)	
#URINARY BLADDER	(25)	(49)	(48)
TRANSITIONAL-CELL CARCINOMA			1 (2%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE A1 (CONTINUED)

	CONTROL (UNTR) 01-0330	LOW DOSE 01-0280	HIGH DOSE 01-0285
ENDOCRINE SYSTEM			
#PITUITARY	(22)	(44)	(44)
CARCINOMA,NOS			1 (2%)
ADENOMA, NOS	2 (9%)	1 (2%)	1 (2%)
CHROMOPHOBE ADENOMA		3 (7%)	7 (16%)
ACIDOPHIL ADENOMA			1 (2%)
ACIDOPHIL CARCINOMA			1 (2%)
BASOPHIL ADENOMA		3 (7%)	2 (5%)
INTERSTITIAL-CELL TUMOR, METASTA			1 (2%)
#ADRENAL	(24)	(48)	(48)
CORTICAL ADENOMA		1 (2%)	
PHEOCHROMOCYTOMA	1 (4%)	3 (6%)	5 (10%)
PHEOCHROMOCYTOMA, MALIGNANT	1 (4%)	1 (2%)	
NEUROBLASTOMA			1 (2%)
#THYROID	(21)	(47)	(47)
FOLLICULAR-CELL ADENOMA	1 (5%)		1 (2%)
C-CELL ADENOMA		2 (4%)	5 (11%)
C-CELL CARCINOMA	2 (10%)	3 (6%)	3 (6%)
SEBACEOUS ADENOCARCINOMA, METAST		1 (2%)	
PAPILLARY CYSTADENOCARCINOMA,NOS			1 (2%)
#PARATHYROID	(13)	(24)	(28)
ADENOMA, NOS			1 (4%)
#PANCREATIC ISLETS	(25)	(48)	(45)
ISLET-CELL ADENOMA	1 (4%)	1 (2%)	4 (9%)
ISLET-CELL CARCINOMA		1 (2%)	1 (2%)
REPRODUCTIVE SYSTEM			
*MAMMARY GLAND	(25)	(49)	(50)
ADENOCARCINOMA, NOS			1 (2%)
FIBROADENOMA		1 (2%)	
*PREPUTIAL GLAND	(25)	(49)	(50)
CARCINOMA,NOS			1 (2%)
ADENOMA, NOS			1 (2%)
#TESTIS	(25)	(49)	(49)
INTERSTITIAL-CELL TUMOR	21 (84%)	44 (90%)	45 (92%)
INTERSTITIAL-CELL TUMOR, MALIGNA			1 (2%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE A1 (CONTINUED)

	CONTROL (UNTR) 01-0330	LOW DOSE 01-0280	HIGH DOSE 01-0285
NERVOUS SYSTEM			
#BRAIN	(25)	(49)	(47)
CARCINOMA, NOS, METASTATIC			1 (2%)
GLIOMA, NOS		1 (2%)	1 (2%)
#CEREBELLUM	(25)	(49)	(47)
GLIOMA, NOS		1 (2%)	
SPECIAL SENSE ORGANS			
*EAR CANAL	(25)	(49)	(50)
SQUAMOUS CELL CARCINOMA	1 (4%)		
*ZYMBAL'S GLAND	(25)	(49)	(50)
SEBACEOUS ADENOCARCINOMA			1 (2%)
MUSCULOSKELETAL SYSTEM			
NONE			
BODY CAVITIES			
*BODY CAVITIES	(25)	(49)	(50)
MESOTHELIOMA, NOS		1 (2%)	
MESOTHELIOMA, MALIGNANT			1 (2%)
*ABDOMINAL CAVITY	(25)	(49)	(50)
OSTEOSARCOMA			1 (2%)
ALL OTHER SYSTEMS			
TAIL			
SQUAMOUS CELL PAPILLOMA			1

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE A1 (CONCLUDED)

	CONTROL (UNTR) 01-0330	LOW DOSE 01-0280	HIGH DOSE 01-0285
ANIMAL DISPOSITION SUMMARY			
ANIMALS INITIALLY IN STUDY	25	50	50
NATURAL DEATH@	5	5	8
MORIBUND SACRIFICE	3	5	5
SCHEDULED SACRIFICE			
ACCIDENTALLY KILLED			
TERMINAL SACRIFICE	17	40	37
ANIMAL MISSING			

@ INCLUDES AUTOLYZED ANIMALS

	CONTROL (UNTR) 01-0330	LOW DOSE 01-0280	HIGH DOSE 01-0285
TUMOR SUMMARY			
TOTAL ANIMALS WITH PRIMARY TUMORS*	21	47	49
TOTAL PRIMARY TUMORS	35	96	119
TOTAL ANIMALS WITH BENIGN TUMORS	21	46	49
TOTAL BENIGN TUMORS	29	67	87
TOTAL ANIMALS WITH MALIGNANT TUMORS	5	19	24
TOTAL MALIGNANT TUMORS	5	25	30
TOTAL ANIMALS WITH SECONDARY TUMORS#	2	1	5
TOTAL SECONDARY TUMORS	2	4	5
TOTAL ANIMALS WITH TUMORS UNCERTAIN- BENIGN OR MALIGNANT	1	4	2
TOTAL UNCERTAIN TUMORS	1	4	2
TOTAL ANIMALS WITH TUMORS UNCERTAIN- PRIMARY OR METASTATIC			
TOTAL UNCERTAIN TUMORS			

* PRIMARY TUMORS: ALL TUMORS EXCEPT SECONDARY TUMORS

SECONDARY TUMORS: METASTATIC TUMORS OR TUMORS INVASIVE INTO AN ADJACENT ORGAN

TABLE A2
SUMMARY OF THE INCIDENCE OF NEOPLASMS IN FEMALE RATS TREATED WITH 1,5-NAPHTHALENEDIAMINE

	CONTROL (UNTR) 02-0330	LOW DOSE 02-0280	HIGH DOSE 02-0285
ANIMALS INITIALLY IN STUDY	25	50	50
ANIMALS NECROPSIED	24	50	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY**	24	50	50
INTEGUMENTARY SYSTEM			
*SKIN	(24)	(50)	(50)
SQUAMOUS CELL CARCINOMA			2 (4%)
RESPIRATORY SYSTEM			
#TRACHEA	(23)	(16)	(10)
PAPILLOMA, NOS			1 (10%)
#LUNG	(24)	(50)	(50)
ALVEOLAR/BRONCHIOLAR ADENOMA		1 (2%)	1 (2%)
ALVEOLAR/BRONCHIOLAR CARCINOMA			1 (2%)
CORTICAL CARCINOMA, METASTATIC		1 (2%)	
C-CELL CARCINOMA, METASTATIC		1 (2%)	
ENDOMETRIAL STROMAL SARCOMA, MET		1 (2%)	
HEMATOPOIETIC SYSTEM			
*MULTIPLE ORGANS	(24)	(50)	(50)
MALIG.LYMPHOMA, HISTIOCYTIC TYPE	1 (4%)		
LEUKEMIA,NOS		1 (2%)	
UNDIFFERENTIATED LEUKEMIA		6 (12%)	1 (2%)
MYELOMONOCYTIC LEUKEMIA	2 (8%)		
CIRCULATORY SYSTEM			
NONE			
DIGESTIVE SYSTEM			
#LIVER	(24)	(50)	(49)
NEOPLASTIC NODULE		3 (6%)	4 (8%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE A2 (CONTINUED)

	CONTROL(UNTR) 02-0330	LOW DOSE 02-0280	HIGH DOSE 02-0285
HEPATOCELLULAR CARCINOMA		1 (2%)	
ENDOMETRIAL STROMAL SARCOMA, INV		1 (2%)	
#STOMACH	(24)	(50)	(49)
CARCINOMA,NOS			1 (2%)
SQUAMOUS CELL PAPILLOMA			1 (2%)
ENDOMETRIAL STROMAL SARCOMA, INV		1 (2%)	
URINARY SYSTEM			
#KIDNEY	(24)	(50)	(49)
LIPOMA		1 (2%)	1 (2%)
#KIDNEY/PELVIS	(24)	(50)	(49)
TRANSITIONAL-CELL PAPILLOMA		1 (2%)	1 (2%)
#URINARY BLADDER	(24)	(48)	(49)
TRANSITIONAL-CELL PAPILLOMA	1 (4%)		
TRANSITIONAL-CELL CARCINOMA			1 (2%)
ENDOCRINE SYSTEM			
#PITUITARY	(21)	(50)	(47)
CARCINOMA,NOS			1 (2%)
ADENOMA, NOS	6 (29%)	1 (2%)	1 (2%)
CHROMOPHOBE ADENOMA		7 (14%)	16 (34%)
CHROMOPHOBE CARCINOMA		1 (2%)	
ACIDOPHIL ADENOMA		1 (2%)	
BASOPHIL ADENOMA		1 (2%)	
PAPILLARY CYSTADENOCARCINOMA,MET			1 (2%)
#ADRENAL	(24)	(50)	(49)
CORTICAL ADENOMA		2 (4%)	1 (2%)
CORTICAL CARCINOMA		1 (2%)	
PHEOCHROMOCYTOMA	1 (4%)		3 (6%)
LIPOMA		1 (2%)	
#THYROID	(21)	(49)	(48)
PAPILLARY CARCINOMA			1 (2%)
FOLLICULAR-CELL CARCINOMA	1 (5%)		1 (2%)
C-CELL ADENOMA		7 (14%)	3 (6%)
C-CELL CARCINOMA	1 (5%)	5 (10%)	1 (2%)
PAPILLARY CYSTADENOMA, NOS		1 (2%)	1 (2%)
PAPILLARY CYSTADENOCARCINOMA,NOS		1 (2%)	2 (4%)

# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE A2 (CONTINUED)

	CONTROL (UNTR) 02-0330	LOW DOSE 02-0280	HIGH DOSE 02-0285
#PANCREATIC ISLETS	(22)	(49)	(47)
ISLET-CELL CARCINOMA	1 (5%)		
REPRODUCTIVE SYSTEM			
*MAMMARY GLAND	(24)	(50)	(50)
ADENOMA, NOS		1 (2%)	1 (2%)
ADENOCARCINOMA, NOS			1 (2%)
PAPILLARY ADENOCARCINOMA		1 (2%)	1 (2%)
INTRADUCTAL PAPILLOMA			1 (2%)
FIBROADENOMA	4 (17%)	5 (10%)	13 (26%)
*CLITORAL GLAND	(24)	(50)	(50)
CARCINOMA,NOS	1 (4%)	3 (6%)	8 (16%)
ADENOMA, NOS			5 (10%)
#UTERUS	(24)	(49)	(48)
ADENOCARCINOMA, NOS	1 (4%)		1 (2%)
ENDOMETRIAL STROMAL POLYP	2 (8%)	14 (29%)	20 (42%)
ENDOMETRIAL STROMAL SARCOMA	1 (4%)	2 (4%)	2 (4%)
#UTERUS/ENDOMETRIUM	(24)	(49)	(48)
ADENOCARCINOMA, NOS		2 (4%)	3 (6%)
#OVARY	(24)	(49)	(49)
GRANULOSA-CELL TUMOR		1 (2%)	
SERTOLI-CELL TUMOR		1 (2%)	
NERVOUS SYSTEM			
#BRAIN	(23)	(50)	(50)
CHROMOPHOBE CARCINOMA, INVASIVE		1 (2%)	
GLIOMA, NOS		1 (2%)	1 (2%)
SPECIAL SENSE ORGANS			
*HARDERIAN GLAND	(24)	(50)	(50)
ADENOCARCINOMA, NOS			1 (2%)
*EAR CANAL	(24)	(50)	(50)
SQUAMOUS CELL CARCINOMA	1 (4%)		
*ZYMBAL'S GLAND	(24)	(50)	(50)
SEBACEOUS ADENOCARCINOMA			3 (6%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

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TABLE A2 (CONTINUED)

	CONTROL (UNTR) 02-0330	LOW DOSE 02-0280	HIGH DOSE 02-0285
MUSCULOSKELETAL SYSTEM			
NONE			
BODY CAVITIES			
*BODY CAVITIES	(24)	(50)	(50)
MESOTHELIOMA, MALIGNANT		1 (2%)	
*ABDOMINAL CAVITY	(24)	(50)	(50)
LEIOMYOSARCOMA	1 (4%)		
*PERITONEUM	(24)	(50)	(50)
ENDOMETRIAL STROMAL SARCOMA, MET		1 (2%)	
ALL OTHER SYSTEMS			
TAIL			
SQUAMOUS CELL PAPILLOMA		1	
DIAPHRAGM			
ENDOMETRIAL STROMAL SARCOMA, MET		1	
ANIMAL DISPOSITION SUMMARY			
ANIMALS INITIALLY IN STUDY	25	50	50
NATURAL DEATH@	4	5	5
MORIBUND SACRIFICE	5	7	7
SCHEDULED SACRIFICE			
ACCIDENTALLY KILLED			
TERMINAL SACRIFICE	16	38	38
ANIMAL MISSING			

@ INCLUDES AUTOLYZED ANIMALS

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE A2 (CONTINUED)

	CONTROL (UNTR) 02-0330	LOW DOSE 02-0260	HIGH DOSE 02-0285
TUMOR SUMMARY			
TOTAL ANIMALS WITH PRIMARY TUMORS*	17	41	49
TOTAL PRIMARY TUMORS	25	76	107
TOTAL ANIMALS WITH BENIGN TUMORS	10	33	44
TOTAL BENIGN TUMORS	14	46	70
TOTAL ANIMALS WITH MALIGNANT TUMORS	10	22	25
TOTAL MALIGNANT TUMORS	11	26	33
TOTAL ANIMALS WITH SECONDARY TUMORS#		5	1
TOTAL SECONDARY TUMORS		8	1
TOTAL ANIMALS WITH TUMORS UNCERTAIN-BENIGN OR MALIGNANT		4	4
TOTAL UNCERTAIN TUMORS		4	4
TOTAL ANIMALS WITH TUMORS UNCERTAIN-PRIMARY OR METASTATIC			
TOTAL UNCERTAIN TUMORS			

* PRIMARY TUMORS: ALL TUMORS EXCEPT SECONDARY TUMORS

SECONDARY TUMORS: METASTATIC TUMORS OR TUMORS INVASIVE INTO AN ADJACENT ORGAN

APPENDIX B

SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MICE TREATED WITH 1,5-NAPHTHALENEDIAMINE

TABLE B1
SUMMARY OF THE INCIDENCE OF NEOPLASMS IN MALE MICE TREATED WITH 1,5-NAPHTHALENEDIAMINE

	CONTROL (UNTR) 05-0330	LOW DOSE 05-0285	HIGH DOSE 05-0290
ANIMALS INITIALLY IN STUDY	50	50	50
ANIMALS MISSING	2	1	
ANIMALS NECROPSIED	39	47	49
ANIMALS EXAMINED HISTOPATHOLOGICALLY**	39	47	46
INTEGUMENTARY SYSTEM			
*SUBCUT TISSUE	(39)	(47)	(49)
FIBROUS HISTIOCYTOMA		1 (2%)	
RESPIRATORY SYSTEM			
#LUNG	(39)	(46)	(45)
HEPATOCELLULAR CARCINOMA, METAST	2 (5%)	2 (4%)	
ALVEOLAR/BRONCHIOLAR ADENOMA	2 (5%)	6 (13%)	2 (4%)
ALVEOLAR/BRONCHIOLAR CARCINOMA	2 (5%)	3 (7%)	
C-CELL CARCINOMA, METASTATIC			1 (2%)
HEMATOPOIETIC SYSTEM			
*MULTIPLE ORGANS	(39)	(47)	(49)
MALIGNANT LYMPHOMA, NOS	11 (28%)	1 (2%)	1 (2%)
MALIG.LYMPHOMA, LYMPHOCYTIC TYPE		3 (6%)	
MALIG.LYMPHOMA, HISTIOCYTIC TYPE		3 (6%)	
MALIGNANT LYMPHOMA, MIXED TYPE		5 (11%)	1 (2%)
#SPLEEN	(38)	(45)	(41)
HEMANGIOSARCOMA		1 (2%)	
MALIGNANT LYMPHOMA, NOS	1 (3%)		
MALIGNANT LYMPHOMA, MIXED TYPE			2 (5%)
#MESENTERIC L. NODE	(36)	(43)	(35)
LYMPHANGIOMA			1 (3%)
MALIG.LYMPHOMA, HISTIOCYTIC TYPE	1 (3%)		
MALIGNANT LYMPHOMA, MIXED TYPE		2 (5%)	
#THYMUS	(13)	(29)	(16)
MALIGNANT LYMPHOMA, NOS			1 (6%)
CIRCULATORY SYSTEM			
NONE			

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE B1 (CONTINUED)

	CONTROL (UNTR) 05-0330	LOW DOSE 05-0285	HIGH DOSE 05-0290
DIGESTIVE SYSTEM			
#LIVER	(39)	(45)	(43)
HEPATOCELLULAR ADENOMA		3 (7%)	6 (14%)
HEPATOCELLULAR CARCINOMA	12 (31%)	10 (22%)	7 (16%)
URINARY SYSTEM			
#KIDNEY	(39)	(47)	(45)
TUBULAR-CELL ADENOMA		1 (2%)	
#URINARY BLADDER	(37)	(45)	(41)
TRANSITIONAL-CELL PAPILLOMA			1 (2%)
*URETHRA	(39)	(47)	(49)
TRANSITIONAL-CELL PAPILLOMA			1 (2%)
ENDOCRINE SYSTEM			
#ADRENAL	(36)	(42)	(42)
PHEOCHROMOCYTOMA, MALIGNANT		1 (2%)	
#THYROID	(38)	(46)	(43)
PAPILLARY CARCINOMA		1 (2%)	1 (2%)
PAPILLARY ADENOMA			1 (2%)
PAPILLARY ADENOCARCINOMA		1 (2%)	
FOLLICULAR-CELL ADENOMA		7 (15%)	14 (33%)
FOLLICULAR-CELL CARCINOMA		1 (2%)	1 (2%)
C-CELL ADENOMA		2 (4%)	
C-CELL CARCINOMA			4 (9%)
PAPILLARY CYSTADENOMA, NOS		1 (2%)	2 (5%)
REPRODUCTIVE SYSTEM			
NONE			
NERVOUS SYSTEM			
NONE			
SPECIAL SENSE ORGANS			
NONE			

# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY

* NUMBER OF ANIMALS NECROPSIED

TABLE B1 (CONTINUED)

	CONTROL (UNTR) 05-0330	LOW DOSE 05-0285	HIGH DOSE 05-0290
MUSCULOSKELETAL SYSTEM			
NONE			
BODY CAVITIES			
*BODY CAVITIES	(39)	(47)	(49)
MESOTHELIOMA, NOS		1 (2%)	
ALL OTHER SYSTEMS			
NONE			
ANIMAL DISPOSITION SUMMARY			
ANIMALS INITIALLY IN STUDY	50	50	50
NATURAL DEATH@	13	9	17
MORIBUND SACRIFICE	2	1	4
SCHEDULED SACRIFICE			
ACCIDENTALLY KILLED			
TERMINAL SACRIFICE	33	39	29
ANIMAL MISSING	2	1	

@ INCLUDES AUTOLYZED ANIMALS

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE B1 (CONCLUDED)

	CONTROL (UNTR) 05-0330	LOW DOSE 05-0285	HIGH DOSE 05-0290
TUMOR SUMMARY			
TOTAL ANIMALS WITH PRIMARY TUMORS*	24	40	25
TOTAL PRIMARY TUMORS	29	54	46
TOTAL ANIMALS WITH BENIGN TUMORS	2	13	20
TOTAL BENIGN TUMORS	2	21	28
TOTAL ANIMALS WITH MALIGNANT TUMORS	22	25	12
TOTAL MALIGNANT TUMORS	27	32	18
TOTAL ANIMALS WITH SECONDARY TUMORS#	2	2	1
TOTAL SECONDARY TUMORS	2	2	1
TOTAL ANIMALS WITH TUMORS UNCERTAIN- BENIGN OR MALIGNANT		1	
TOTAL UNCERTAIN TUMORS		1	
TOTAL ANIMALS WITH TUMORS UNCERTAIN- PRIMARY OR METASTATIC			
TOTAL UNCERTAIN TUMORS			

* PRIMARY TUMORS: ALL TUMORS EXCEPT SECONDARY TUMORS

# SECONDARY TUMORS: METASTATIC TUMORS OR TUMORS INVASIVE INTO AN ADJACENT ORGAN

TABLE B2
SUMMARY OF THE INCIDENCE OF NEOPLASMS IN FEMALE MICE TREATED WITH 1,5-NAPHTHALENEDIAMINE

	CONTROL (UNTR) 06-0330	LOW DOSE 06-0285	HIGH DOSE 06-0290
ANIMALS INITIALLY IN STUDY	50	50	50
ANIMALS MISSING			1
ANIMALS NECROPSIED	49	50	46
ANIMALS EXAMINED HISTOPATHOLOGICALLY**	49	49	46
INTEGUMENTARY SYSTEM			
*SKIN	(49)	(50)	(46)
UNDIFFERENTIATED CARCINOMA			1 (2%)
*SUBCUT TISSUE	(49)	(50)	(46)
MIXED MESENCHYMAL TUMOR, MALIGNA			1 (2%)
RESPIRATORY SYSTEM			
#LUNG	(49)	(48)	(46)
UNDIFFERENTIATED CARCINOMA METAS			1 (2%)
HEPATOCELLULAR CARCINOMA, METAST		2 (4%)	
ALVEOLAR/BRONCHIOLAR ADENOMA		9 (19%)	2 (4%)
ALVEOLAR/BRONCHIOLAR CARCINOMA		1 (2%)	3 (7%)
HEMATOPOIETIC SYSTEM			
*MULTIPLE ORGANS	(49)	(50)	(46)
MALIGNANT LYMPHOMA, NOS	10 (20%)	1 (2%)	
MALIG.LYMPHOMA, LYMPHOCYTIC TYPE		1 (2%)	
MALIG.LYMPHOMA, HISTIOCYTIC TYPE	1 (2%)		
MALIGNANT LYMPHOMA, MIXED TYPE		10 (20%)	3 (7%)
LYMPHOCYTIC LEUKEMIA		1 (2%)	
#BONE MARROW	(40)	(48)	(45)
HEPATOCELLULAR CARCINOMA, METAST		1 (2%)	
#SPLEEN	(45)	(48)	(45)
HEPATOCELLULAR CARCINOMA, METAST		1 (2%)	
HEMANGIOSARCOMA	1 (2%)		
MALIGNANT LYMPHOMA, MIXED TYPE		1 (2%)	
#MEDIASTINAL L.NODE	(44)	(45)	(40)
MALIGNANT LYMPHOMA, MIXED TYPE		1 (2%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE B2 (CONTINUED)

	CONTROL(UNTR) 06-0330	LOW DOSE 06-0285	HIGH DOSE 06-0290
#PANCREATIC L.NODE	(44)	(45)	(49)
MALIGNANT LYMPHOMA, NOS	1 (2%)		
#MESENTERIC L. NODE	(44)	(45)	(50)
HEPATOCELLULAR CARCINOMA, METAST		2 (4%)	
MALIGNANT LYMPHOMA, NOS		1 (2%)	
MALIGNANT LYMPHOMA, MIXED TYPE		1 (2%)	1 (2%)
#LIVER	(46)	(49)	(46)
MALIGNANT LYMPHOMA, NOS	1 (2%)		
#DUODENUM	(42)	(47)	(48)
MALIGNANT LYMPHOMA, MIXED TYPE			1 (2%)
#KIDNEY	(45)	(49)	(46)
MALIGNANT LYMPHOMA, MIXED TYPE		2 (4%)	
CIRCULATORY SYSTEM			
#HEART	(43)	(49)	(46)
UNDIFFERENTIATED CARCINOMA, METAS			1 (2%)
DIGESTIVE SYSTEM			
#LIVER	(46)	(49)	(46)
HEPATOCELLULAR ADENOMA		3 (6%)	11 (24%)
HEPATOCELLULAR CARCINOMA	1 (2%)	25 (51%)	16 (35%)
#PANCREAS	(38)	(47)	(44)
HEPATOCELLULAR CARCINOMA, METAST		1 (2%)	
#STOMACH	(41)	(47)	(36)
SQUAMOUS CELL PAPILLOMA		3 (6%)	
HEPATOCELLULAR CARCINOMA, METAST		2 (4%)	
URINARY SYSTEM			
#URINARY BLADDER	(39)	(46)	(48)
TRANSITIONAL-CELL PAPILLOMA			1 (2%)
ENDOCRINE SYSTEM			
#PITUITARY	(32)	(35)	(37)
ADENOMA, NOS	2 (6%)		1 (3%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE B2 (CONTINUED)

	CONTROL (UNTR) 06-0331	LOW DOSE 06-0285	HIGH DOSE 06-0290
CHROMOPHOBE ADENOMA		3 (9%)	
ACIDOPHIL ADENOMA		1 (3%)	
#ADRENAL	(46)	(44)	(41)
PHEOCHROMOCYTOMA	3 (7%)		
PHEOCHROMOCYTOMA, MALIGNANT	1 (2%)		
#THYROID	(44)	(43)	(45)
PAPILLARY ADENOMA		1 (2%)	2 (4%)
FOLLICULAR-CELL ADENOMA	2 (5%)	6 (14%)	10 (22%)
FOLLICULAR-CELL CARCINOMA	2 (5%)		1 (2%)
C-CELL ADENOMA		1 (2%)	2 (4%)
C-CELL CARCINOMA		1 (2%)	6 (13%)
PAPILLARY CYSTADENOMA, NOS		8 (19%)	4 (9%)
REPRODUCTIVE SYSTEM			
*MAMMARY GLAND	(49)	(50)	(46)
ACINAR-CELL CARCINOMA			1 (2%)
#UTERUS	(44)	(45)	(43)
ENDOMETRIAL STROMAL POLYP	1 (2%)		1 (2%)
HEMANGIOMA		2 (4%)	1 (2%)
#OVARY	(44)	(45)	(41)
HEPATOCELLULAR CARCINOMA, METAST		1 (2%)	
GRANULOSA-CELL TUMOR			1 (2%)
TUBULAR ADENOMA	1 (2%)	2 (4%)	
NERVOUS SYSTEM			
NONE			
SPECIAL SENSE ORGANS			
*HARDERIAN GLAND	(49)	(50)	(46)
CYSTADENOMA, NOS		1 (2%)	
MUSCULOSKELETAL SYSTEM			
NONE			

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE B2 (CONCLUDED)

	CONTROL (UNTR) 06-0320	LOW DOSE 06-0285	HIGH DOSE 06-0290
BODY CAVITIES			
*BODY CAVITIES	(49)	(50)	(46)
MESOTHELIOMA, NOS			1 (2%)
ALL OTHER SYSTEMS			
NONE			
ANIMAL DISPOSITION SUMMARY			
ANIMALS INITIALLY IN STUDY	50	50	50
NATURAL DEATH@	17	9	9
MORIBUND SACRIFICE	3		5
SCHEDULED SACRIFICE			
ACCIDENTALLY KILLED			1
TERMINAL SACRIFICE	30	41	34
ANIMAL MISSING			1
@ INCLUDES AUTOLYZED ANIMALS			
TUMOR SUMMARY			
TOTAL ANIMALS WITH PRIMARY TUMORS*	21	41	37
TOTAL PRIMARY TUMORS	28	88	71
TOTAL ANIMALS WITH BENIGN TUMORS	9	25	23
TOTAL BENIGN TUMORS	10	42	35
TOTAL ANIMALS WITH MALIGNANT TUMORS	16	37	27
TOTAL MALIGNANT TUMORS	18	46	34
TOTAL ANIMALS WITH SECONDARY TUMORS#		3	1
TOTAL SECONDARY TUMORS		10	2
TOTAL ANIMALS WITH TUMORS UNCERTAIN-BENIGN OR MALIGNANT			2
TOTAL UNCERTAIN TUMORS			2
TOTAL ANIMALS WITH TUMORS UNCERTAIN-PRIMARY OR METASTATIC			
TOTAL UNCERTAIN TUMORS			

* PRIMARY TUMORS: ALL TUMORS EXCEPT SECONDARY TUMORS

# SECONDARY TUMORS: METASTATIC TUMORS OR TUMORS INVASIVE INTO AN ADJACENT ORGAN

APPENDIX C

SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN RATS TREATED WITH 1,5-NAPHTHALENEDIAMINE

TABLE C1
SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MALE RATS TREATED WITH 1,5-NAPHTHALENEDIAMINE

	CONTROL (UNTR) 01-0330	LOW DOSE 01-0282	HIGH DOSE 01-0285
ANIMALS INITIALLY IN STUDY	25	50	50
ANIMALS NECROPSIED	25	49	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY **	25	49	49
INTEGUMENTARY SYSTEM			
*SKIN	(25)	(49)	(50)
VEGETABLE FOREIGN BODY		1 (2%)	
EPIDERMAL INCLUSION CYST		1 (2%)	
ABSCESS, NOS		1 (2%)	
SCAR	1 (4%)		
*SUBCUT TISSUE	(25)	(49)	(50)
CYST, NOS			1 (2%)
RESPIRATORY SYSTEM			
*LARYNX	(25)	(49)	(50)
INFLAMMATION, ACUTE/CHRONIC		1 (2%)	
INFLAMMATION, CHRONIC		11 (22%)	1 (2%)
#LUNG/BRONCHIOLE	(25)	(49)	(47)
METAPLASIA, NOS		2 (4%)	
#LUNG	(25)	(49)	(47)
BRONCHIECTASIS		4 (8%)	3 (6%)
BRONCHOPNEUMONIA, NOS			1 (2%)
BRONCHOPNEUMONIA NECROTIZING	1 (4%)		
BRONCHOPNEUMONIA, ACUTE			2 (4%)
ABSCESS, NOS		1 (2%)	
PNEUMONIA, CHRONIC MURINE		22 (45%)	5 (11%)
HEMATOPOIETIC SYSTEM			
#BONE MARROW	(23)	(49)	(47)
HYPERPLASIA, NOS		3 (6%)	3 (6%)
HYPERPLASIA, HEMATOPOIETIC			2 (4%)
#SPLEEN	(25)	(49)	(48)
CONGESTION, NOS	1 (4%)	1 (2%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE C1 (CONTINUED)

	CONTROL(UNTR) 01-0330	LOW DOSE 01-0280	HIGH DOSE 01-0285
HEMOSIDEROSIS		2 (4%)	
#MANDIBULAR L. NODE	(24)	(47)	(47)
INFLAMMATION, CHRONIC			1 (2%)
HYPERPLASIA, PLASMA CELL	2 (8%)		
#THYMUS	(13)	(35)	(36)
CYST, NOS		1 (3%)	
INFLAMMATION, CHRONIC		1 (3%)	
CIRCULATORY SYSTEM			
#HEART	(25)	(49)	(47)
PERIARTERITIS		1 (2%)	
DEGENERATION, NOS		25 (51%)	7 (15%)
#MYOCARDIUM	(25)	(49)	(47)
INFLAMMATION, CHRONIC	1 (4%)		
DIGESTIVE SYSTEM			
#LIVER	(25)	(49)	(49)
CONGESTION, CHRONIC PASSIVE		1 (2%)	2 (4%)
CHOLANGIOFIBROSIS		4 (8%)	1 (2%)
DEGENERATION, NOS	1 (4%)		
NECROSIS, FOCAL		1 (2%)	1 (2%)
METAMORPHOSIS FATTY	1 (4%)	1 (2%)	
BASOPHILIC CYTO CHANGE	1 (4%)	3 (6%)	3 (6%)
FOCAL CELLULAR CHANGE		1 (2%)	
CLEAR-CELL CHANGE			1 (2%)
HYPERPLASIA, FOCAL	1 (4%)		
ANGIECTASIS	1 (4%)		
#LIVER/CENTRILOBULAR	(25)	(49)	(49)
NECROSIS, NOS			1 (2%)
#PANCREAS	(25)	(48)	(45)
FIBROSIS, FOCAL			1 (2%)
ATROPHY, FOCAL		1 (2%)	
#STOMACH	(24)	(47)	(47)
ULCER, ACUTE			1 (2%)
HYPERPLASIA, BASAL CELL		1 (2%)	
HYPERKERATOSIS		1 (2%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C1 (CONTINUED)

	CONTROL (UNTR) 01-0331	LOW DOSE 01-0280	HIGH DOSE 01-0285
#COLON	(23)	(45)	(46)
PARASITISM		5 (11%)	
URINARY SYSTEM			
#KIDNEY	(25)	(49)	(48)
CYST, NOS		1 (2%)	
PYELONEPHRITIS, FOCAL			1 (2%)
PYELONEPHRITIS, CHRONIC		1 (2%)	
NEPHROSIS, NOS	21 (84%)	42 (86%)	15 (31%)
HYPERPLASIA, EPITHELIAL			2 (4%)
#KIDNEY/CORTEX	(25)	(49)	(48)
MULTILOCULAR CYST	1 (4%)		
#KIDNEY/PELVIS	(25)	(49)	(48)
CALCULUS, NOS			1 (2%)
#URINARY BLADDER	(25)	(49)	(48)
CALCULUS, NOS		2 (4%)	1 (2%)
ENDOCRINE SYSTEM			
#PITUITARY	(22)	(44)	(44)
HYPERPLASIA, FOCAL			1 (2%)
#ADRENAL	(24)	(48)	(48)
HYPERPLASIA, NODULAR			1 (2%)
#ADRENAL CORTEX	(24)	(48)	(48)
HYPERPLASIA, NOS			3 (6%)
HYPERPLASIA, FOCAL	1 (4%)		
#ADRENAL MEDULLA	(24)	(48)	(48)
HYPERPLASIA, NOS		1 (2%)	2 (4%)
HYPERPLASIA, FOCAL	4 (17%)		
#THYROID	(21)	(47)	(47)
CYST, NOS		1 (2%)	
FOLLICULAR CYST, NOS		1 (2%)	1 (2%)
INFLAMMATION, CHRONIC			6 (13%)
HYPERPLASIA, C-CELL		1 (2%)	
#PARATHYROID	(13)	(24)	(28)
HYPERPLASIA, NOS			2 (7%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C1 (CONTINUED)

	CONTROL (UNTR) 01-0230	LOW DOSE 01-0280	HIGH DOSE 01-0285
#PANCREATIC ISLETS	(25)	(48)	(45)
HYPERPLASIA, NOS		1 (2%)	
REPRODUCTIVE SYSTEM			
*MAMMARY GLAND	(25)	(49)	(50)
ABSCESS, NOS			1 (2%)
LACTATION		3 (6%)	
*MAMMARY DUCT	(25)	(49)	(50)
HEMORRHAGE	1 (4%)		
#PROSTATE	(25)	(49)	(47)
INFLAMMATION, SUPPURATIVE	2 (8%)		
INFLAMMATION, ACUTE/CHRONIC		3 (6%)	
INFLAMMATION, CHRONIC		1 (2%)	
ATROPHY, NOS		10 (20%)	17 (36%)
*SEMINAL VESICLE	(25)	(49)	(50)
ATROPHY, NOS		10 (20%)	17 (34%)
#TESTIS	(25)	(49)	(49)
PERIARTERITIS			1 (2%)
ATROPHY, NOS	5 (20%)	10 (20%)	
ATROPHY, FOCAL		3 (6%)	
SPERMATOGENIC ARREST		1 (2%)	
HYPOSPERMATOGENESIS		1 (2%)	
*EPIDIDYMIS	(25)	(49)	(50)
NECROSIS, NOS		1 (2%)	
NERVOUS SYSTEM			
#CEREBRUM	(25)	(49)	(47)
HEMORRHAGE		1 (2%)	
#BRAIN	(25)	(49)	(47)
HYDROCEPHALUS, NOS		2 (4%)	1 (2%)
#BRAIN STEM	(25)	(49)	(47)
HEMORRHAGE		1 (2%)	
SPECIAL SENSE ORGANS			
NONE			

# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C1 (CONCLUDED)

	CONTROL(UNTR) 01-0330	LOW DOSE 01-0280	HIGH DOSE 01-0285
MUSCULOSKELETAL SYSTEM			
NONE			
BODY CAVITIES			
*ABDOMINAL CAVITY	(25)	(49)	(50)
NECROSIS, FAT		1 (2%)	2 (4%)
ALL OTHER SYSTEMS			
NONE			
SPECIAL MORPHOLOGY SUMMARY			
NO LESION REPORTED	4	1	
AUTO/NECROPSY/NO HISTO			1
AUTOLYSIS/NO NECROPSY		1	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C2
SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN FEMALE RATS TREATED WITH 1,5-NAPHTHALENEDIAMINE

	CONTROL(UNTR) 02-0330	LOW DOSE 02-0280	HIGH DOSE 02-0285
ANIMALS INITIALLY IN STUDY	25	50	50
ANIMALS NECROPSIED	24	50	50
ANIMALS EXAMINED HISTOPATHOLOGICALLY**	24	50	50
INTEGUMENTARY SYSTEM			
NONE			
RESPIRATORY SYSTEM			
#LARYNX	(24)	(50)	(50)
INFLAMMATION, CHRONIC		1 (2%)	
#LUNG/BRONCHIOLE	(24)	(50)	(50)
METAPLASIA, NOS		2 (4%)	
#LUNG	(24)	(50)	(50)
BRONCHIECTASIS			2 (4%)
BRONCHOPNEUMONIA, NOS			1 (2%)
INFLAMMATION, INTERSTITIAL		1 (2%)	1 (2%)
PNEUMONIA, CHRONIC MURINE		19 (38%)	2 (4%)
HEMATOPOIETIC SYSTEM			
#BONE MARROW	(21)	(49)	(49)
HYPERPLASIA, NOS		1 (2%)	
HYPERPLASIA, HEMATOPOIETIC		1 (2%)	1 (2%)
#SPLEEN	(23)	(50)	(49)
METAPLASIA, NOS			1 (2%)
HYPERPLASIA, HEMATOPOIETIC		1 (2%)	
HYPERPLASIA, ERYTHROID		1 (2%)	
HEMATOPOIESIS		3 (6%)	1 (2%)
ERYTHROPOIESIS	1 (4%)		
#LYMPH NODE	(19)	(48)	(49)
HYPERPLASIA, LYMPHOID		1 (2%)	
#MANDIBULAR L. NODE	(19)	(48)	(49)
CONGESTION, NOS			1 (2%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE C2 (CONTINUED)

	CONTROL (UNTR) 02-0330	LOW DOSE 02-0280	HIGH DOSE 02-0285
HYPERPLASIA, PLASMA CELL	2 (11%)		
CIRCULATORY SYSTEM			
#HEART	(24)	(50)	(50)
DEGENERATION, NOS		5 (10%)	2 (4%)
#MYOCARDIUM	(24)	(50)	(50)
CALCIFICATION, FOCAL	1 (4%)		
*AORTA	(24)	(50)	(50)
MEDIAL CALCIFICATION	1 (4%)		
*CORONARY ARTERY	(24)	(50)	(50)
MEDIAL CALCIFICATION	1 (4%)		
CALCIFICATION, NOS		1 (2%)	
DIGESTIVE SYSTEM			
#LIVER	(24)	(50)	(49)
CONGESTION, CHRONIC PASSIVE		1 (2%)	
CHOLANGIOFIBROSIS			1 (2%)
NECROSIS, FOCAL		1 (2%)	2 (4%)
INFARCT, FOCAL		1 (2%)	
METAMORPHOSIS FATTY		1 (2%)	
BASOPHILIC CYTO CHANGE	10 (42%)	2 (4%)	12 (24%)
CLEAR-CELL CHANGE		1 (2%)	
ANGIECTASIS	1 (4%)		
HYPERPLASIA, BASOPHILIC	1 (4%)		
#LIVER/CENTRILOBULAR	(24)	(50)	(49)
NECROSIS, NOS		2 (4%)	
#PANCREAS	(22)	(49)	(47)
ATROPHY, FOCAL			1 (2%)
#STOMACH	(24)	(50)	(49)
ATYPIA, NOS		1 (2%)	
HYPERPLASIA, BASAL CELL		2 (4%)	3 (6%)
#COLON	(24)	(47)	(46)
PARASITISM		3 (6%)	
URINARY SYSTEM			
#KIDNEY	(24)	(50)	(49)
CYST, NOS		1 (2%)	

# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C2 (CONTINUED)

	CONTROL (UNTR) 02-0330	LOW DOSE 02-0280	HIGH DOSE 02-0285
GLOMERULONEPHRITIS, NOS		1 (2%)	
NEPHROSIS, NOS	8 (33%)	7 (14%)	1 (2%)
NECROSIS, MEDULLARY			1 (2%)
CALCIFICATION, FOCAL		1 (2%)	
#KIDNEY/TUBULE	(24)	(50)	(49)
CALCIFICATION, NOS	1 (4%)		
ENDOCRINE SYSTEM			
#PITUITARY	(21)	(50)	(47)
CYST, NOS		1 (2%)	1 (2%)
HEMOSIDEROSIS	1 (5%)		
HYPERPLASIA, NOS		1 (2%)	
#ADRENAL CORTEX	(24)	(50)	(49)
HYPERPLASIA, NOS		1 (2%)	3 (6%)
#ADRENAL MEDULLA	(24)	(50)	(49)
HYPERPLASIA, NOS		1 (2%)	
#THYROID	(21)	(49)	(48)
FOLLICULAR CYST, NOS		1 (2%)	
INFLAMMATION, CHRONIC			7 (15%)
HYPERPLASIA, C-CELL	1 (5%)		
REPRODUCTIVE SYSTEM			
*MAMMARY GLAND	(24)	(50)	(50)
GALACTOCELE			3 (6%)
LACTATION		12 (24%)	4 (8%)
*VAGINA	(24)	(50)	(50)
HYPERKERATOSIS		1 (2%)	
#UTERUS	(24)	(49)	(48)
HYDROMETRA		1 (2%)	
EPIDERMAL INCLUSION CYST			1 (2%)
THROMBOSIS, NOS		1 (2%)	
PYOMETRA		6 (12%)	3 (6%)
ATROPHY, NOS		3 (6%)	
#UTERUS/ENDOMETRIUM	(24)	(49)	(48)
CYST, NOS			1 (2%)

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C2 (CONTINUED)

	CONTROL (UNTR) 02-0330	LOW DOSE 02-0280	HIGH DOSE 02-0285
INFLAMMATION, SUPPURATIVE	3 (13%)		
INFLAMMATION, CHRONIC		1 (2%)	
HYPERPLASIA, CYSTIC		4 (8%)	1 (2%)
METAPLASIA, SQUAMOUS	1 (4%)		
#OVARY/OVIDUCT	(24)	(49)	(48)
INFLAMMATION, SUPPURATIVE	5 (21%)		
ABSCESS, NOS			2 (4%)
#OVARY	(24)	(49)	(49)
CYST, NOS	2 (8%)	8 (16%)	
INFLAMMATION, SUPPURATIVE	1 (4%)		
INFLAMMATION, ACUTE		1 (2%)	
ABSCESS, NOS		1 (2%)	
NERVOUS SYSTEM			
#BRAIN	(23)	(50)	(50)
HYDROCEPHALUS, NOS		1 (2%)	
SPECIAL SENSE ORGANS			
*EYE	(24)	(50)	(50)
INFLAMMATION, NOS			1 (2%)
PHTHISIS BULBI			1 (2%)
MUSCULOSKELETAL SYSTEM			
NONE			
BODY CAVITIES			
*ABDOMINAL CAVITY	(24)	(50)	(50)
NECROSIS, FAT		1 (2%)	
ALL OTHER SYSTEMS			
CRANIOBUCCAL POUCH			
CYST, NOS		1	
SPECIAL MORPHOLOGY SUMMARY			
NO LESION REPORTED	1		

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE C2 (CONCLUDED)

	CONTROL (UNTR) 02-0330	LOW DOSE 02-0280	HIGH DOSE 02-0285
AUTOLYSIS/NO NECROPSY	1		

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

APPENDIX D

SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MICE TREATED WITH 1,5-NAPHTHALENEDIAMINE

TABLE D1
SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN MALE MICE TREATED WITH 1,5-NAPHTHALENEDIAMINE

	CONTROL (UNTR) 05-0330	LOW DOSE 05-0285	HIGH DOSE 05-0290
ANIMALS INITIALLY IN STUDY	50	50	50
ANIMALS MISSING	2	1	
ANIMALS NECROPSIED	39	47	49
ANIMALS EXAMINED HISTOPATHOLOGICALLY**	39	47	46
INTEGUMENTARY SYSTEM			
*SKIN	(39)	(47)	(49)
EPIDERMAL INCLUSION CYST	1 (3%)		
INFLAMMATION, CHRONIC	1 (3%)		
FIBROSIS	1 (3%)		
*SUBCUT TISSUE	(39)	(47)	(49)
ABSCESS, NOS		1 (2%)	
RESPIRATORY SYSTEM			
#LUNG	(39)	(46)	(45)
BRONCHOPNEUMONIA, NOS			1 (2%)
HYPERPLASIA, ADENOMATOUS			14 (31%)
HEMATOPOIETIC SYSTEM			
#SPLEEN	(38)	(45)	(41)
HYPERPLASIA, LYMPHOID	1 (3%)		
HEMATOPOIESIS	1 (3%)	2 (4%)	
ERYTHROPOIESIS	3 (8%)		
#MANDIBULAR L. NODE	(36)	(43)	(35)
HYPERPLASIA, PLASMA CELL	1 (3%)		
#MESENTERIC L. NODE	(36)	(43)	(35)
INFLAMMATION, GRANULOMATOUS			1 (3%)
HYPERPLASIA, NOS	4 (11%)	1 (2%)	
HYPERPLASIA, LYMPHOID	4 (11%)		
CIRCULATORY SYSTEM			
#HEART	(39)	(47)	(44)
PERIARTERITIS		1 (2%)	

# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE D1 (CONTINUED)

	CONTROL (UNTR) 05-0330	LOW DOSE 05-0285	HIGH DOSE 05-0290
DIGESTIVE SYSTEM			
#LIVER	(39)	(45)	(43)
NECROSIS, FOCAL		1 (2%)	
CLEAR-CELL CHANGE		1 (2%)	
HYPERPLASIA, FOCAL	3 (8%)		
#PANCREATIC DUCT	(36)	(46)	(40)
DILATATION, NOS		1 (2%)	
#DUODENUM	(37)	(47)	(39)
AMYLOIDOSIS			1 (3%)
#JEJUNUM	(37)	(47)	(39)
AMYLOIDOSIS	1 (3%)		1 (3%)
#ILEUM	(37)	(47)	(39)
AMYLOIDOSIS	2 (5%)		
#COLON	(37)	(40)	(36)
PARASITISM		2 (5%)	
URINARY SYSTEM			
#KIDNEY	(39)	(47)	(45)
HYDRONEPHROSIS		1 (2%)	2 (4%)
PYELONEPHRITIS, FOCAL			1 (2%)
PYELONEPHRITIS, CHRONIC		1 (2%)	4 (9%)
GLOMERULOSCLEROSIS, NOS	3 (8%)		
NECROSIS, MEDULLARY			3 (7%)
INFARCT, HEALED		1 (2%)	
AMYLOIDOSIS		5 (11%)	12 (27%)
CALCIFICATION, NOS		1 (2%)	
CALCIFICATION, FOCAL		1 (2%)	1 (2%)
#KIDNEY/CORTEX	(39)	(47)	(45)
SCAR	1 (3%)		
#RENAL PAPILLA	(39)	(47)	(45)
CALCIFICATION, NOS			5 (11%)
#PERIRENAL TISSUE	(39)	(47)	(45)
ABSCESS, NOS			1 (2%)
#KIDNEY/GLOMERULUS	(39)	(47)	(45)
AMYLOIDOSIS	2 (5%)		

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D1 (CONTINUED)

	CONTROL (UNTR) 05-C330	LOW DOSE 05-0285	HIGH DOSE 05-C290
#URINARY BLADDER	(37)	(45)	(41)
CALCULUS, NOS		1 (2%)	
PERIARTERITIS		2 (4%)	
HYPERPLASIA, EPITHELIAL			1 (2%)
ENDOCRINE SYSTEM			
#ADRENAL	(36)	(42)	(42)
AMYLOIDOSIS	2 (6%)	1 (2%)	
#THYROID	(38)	(46)	(43)
CYSTIC FOLLICLES	1 (3%)		
INFLAMMATION, ACUTE			1 (2%)
INFLAMMATION, ACUTE/CHRONIC			1 (2%)
AMYLOIDOSIS	1 (3%)		
HYPERPLASIA, FOCAL		1 (2%)	
HYPERPLASIA, PAPILLARY		11 (24%)	7 (16%)
HYPERPLASIA, ADENOMATOUS			2 (5%)
HYPERPLASIA, FOLLICULAR-CELL	2 (5%)		
#PARATHYROID	(28)	(10)	(8)
HYPERPLASIA, NOS	1 (4%)		
#PANCREATIC ISLETS	(36)	(46)	(45)
HYPERPLASIA, NOS	1 (3%)		
REPRODUCTIVE SYSTEM			
*CLITORAL GLAND	(39)	(47)	(49)
DILATATION, NOS		1 (2%)	
NERVOUS SYSTEM			
#SUBARACHNOID SPACE	(38)	(47)	(40)
HEMORRHAGE			1 (3%)
#BRAIN	(38)	(47)	(40)
HEMORRHAGE			1 (3%)
SPECIAL SENSE ORGANS			
NONE			

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D1 (CONCLUDED)

	CONTROL (UNTR) 05-0320	LOW DOSE 05-0285	HIGH DOSE 05-0290
MUSCULOSKELETAL SYSTEM			
NONE			
BODY CAVITIES			
NONE			
ALL OTHER SYSTEMS			
*MULTIPLE ORGANS	(39)	(47)	(49)
AMYLOIDOSIS	1 (3%)		
SPECIAL MORPHOLOGY SUMMARY			
NO LESION REPORTED	5	1	1
ANIMAL MISSING/NO NECROPSY	2	1	
NECROPSY PERF/NO HISTO PERFORMED			1
AUTO/NECROPSY/HISTO PERF			3
AUTO/NECROPSY/NO HISTO			2
AUTOLYSIS/NO NECROPSY	9	2	1

# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D2
SUMMARY OF THE INCIDENCE OF NONNEOPLASTIC LESIONS IN FEMALE MICE TREATED WITH 1,5-NAPHTHALENEDIAMINE

	CONTROL (UNTR) 06-0330	LOW DOSE 06-0285	HIGH DOSE 06-0290
ANIMALS INITIALLY IN STUDY	50	50	50
ANIMALS MISSING			1
ANIMALS NECROPSIED	49	50	45
ANIMALS EXAMINED HISTOPATHOLOGICALLY**	49	49	46
INTEGUMENTARY SYSTEM			
*SKIN	(49)	(50)	(46)
INFLAMMATION, ACUTE	1 (2%)		
RESPIRATORY SYSTEM			
#LUNG	(49)	(48)	(46)
BRONCHOPNEUMONIA, NOS			1 (2%)
PNEUMONIA, CHRONIC MURINE		3 (6%)	2 (4%)
HYPERPLASIA, ADENOMATOUS			14 (30%)
METAPLASIA, NOS			2 (4%)
HEMATOPOIETIC SYSTEM			
#BONE MARROW	(40)	(48)	(45)
FIBROSIS		1 (2%)	
HYPERPLASIA, HEMATOPOIETIC		2 (4%)	
#SPLEEN	(45)	(48)	(45)
HEMOSIDEROSIS		1 (2%)	
HYPERPLASIA, LYMPHOID	1 (2%)		
ERYTHROPOIESIS	4 (9%)		
#LYMPH NODE OF THORAX	(44)	(45)	(40)
HYPERPLASIA, NOS	1 (2%)		
#PANCREATIC L.NODE	(44)	(45)	(40)
HEMATOPOIESIS	1 (2%)		
#LUMBAR LYMPH NODE	(44)	(45)	(40)
HYPERPLASIA, NOS	1 (2%)		
#MESENTERIC L. NODE	(44)	(45)	(40)
HYPERPLASIA, NOS	1 (2%)		

# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED
**EXCLUDES PARTIALLY AUTOLYZED ANIMALS

TABLE D2 (CONTINUED)

	CONTROL(UNTR) 06-0330	LOW DOSE 06-0285	HIGH DOSE 06-0290
HYPERPLASIA, LYMPHOID	1 (2%)		
HEMATOPOIESIS	1 (2%)		
#RENAL LYMPH NODE	(44)	(45)	(40)
HEMATOPOIESIS	1 (2%)		
CIRCULATORY SYSTEM			
#HEART	(49)	(49)	(46)
CALCIFICATION, FOCAL			1 (2%)
DIGESTIVE SYSTEM			
#LIVER	(46)	(49)	(46)
INFARCT, NOS		1 (2%)	
CLEAR-CELL CHANGE		1 (2%)	
HYPERPLASTIC NODULE	1 (2%)		
HEMATOPOIESIS	1 (2%)		
#PANCREAS	(38)	(47)	(44)
PERIARTERITIS			1 (2%)
ATROPHY, NOS			1 (2%)
#JEJUNUM	(42)	(47)	(45)
AMYLOIDOSIS			1 (2%)
#ILEUM	(42)	(47)	(45)
AMYLOIDOSIS		2 (4%)	
URINARY SYSTEM			
#KIDNEY	(46)	(49)	(46)
GLOMERULONEPHRITIS, NOS		1 (2%)	
GLOMERULOSCLEROSIS, NOS	2 (4%)		
AMYLOIDOSIS		3 (6%)	1 (2%)
#URINARY BLADDER	(43)	(46)	(44)
HYPERPLASIA, EPITHELIAL			4 (9%)
DYSPLASIA, NOS			1 (2%)
ENDOCRINE SYSTEM			
#PITUITARY	(34)	(35)	(30)
HYPERPLASIA, FOCAL		1 (3%)	

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D2 (CONTINUED)

	CONTROL (UNTR) 06-0330	LOW DOSE 06-0285	HIGH DOSE 06-0290
#ADRENAL	(46)	(44)	(44)
THROMBUS, ORGANIZED			1 (2%)
AMYLOIDOSIS		1 (2%)	
#THYROID	(44)	(49)	(45)
INFLAMMATION, ACUTE		1 (2%)	1 (2%)
INFLAMMATION, ACUTE FOCAL	1 (2%)		
HYPERPLASIA, NOS			2 (4%)
HYPERPLASIA, PAPILLARY		1 (2%)	2 (4%)
HYPERPLASIA, FOLLICULAR-CELL	2 (5%)		
#PARATHYROID	(24)	(12)	(15)
HYPERPLASIA, NOS			1 (7%)
REPRODUCTIVE SYSTEM			
#UTERUS	(44)	(45)	(43)
HYDROMETRA			5 (12%)
#UTERUS/ENDOMETRIUM	(44)	(45)	(43)
HYPERPLASIA, CYSTIC	30 (68%)	5 (11%)	
#OVARY/OVIDUCT	(44)	(45)	(43)
ABSCESS, NOS			1 (2%)
#OVARY	(44)	(45)	(41)
CYST, NOS	7 (16%)	3 (7%)	3 (7%)
HEMORRHAGIC CYST	4 (9%)		
ABSCESS, NOS	1 (2%)		
INFLAMMATION, CHRONIC	2 (5%)		
NERVOUS SYSTEM			
#BRAIN/MENINGES	(46)	(47)	(43)
INFLAMMATION, CHRONIC			1 (2%)
#BRAIN	(46)	(47)	(43)
PERIARTERITIS			1 (2%)
SPECIAL SENSE ORGANS			
NONE			

# NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

TABLE D2 (CONCLUDED)

	CONTROL (UNTR) 06-0330	LOW DOSE 06-0265	HIGH DOSE 06-0290
MUSCULOSKELETAL SYSTEM			
*SKELETAL MUSCLE	(49)	(50)	(46)
ABSCESS, NOS	1 (2%)		
BODY CAVITIES			
*ABDOMINAL CAVITY	(49)	(50)	(46)
NECROSIS, FAT		1 (2%)	
ALL OTHER SYSTEMS			
ADIPOSE TISSUE			
STEATITIS	1		
NECROSIS, FAT	1		
SPECIAL MORPHOLOGY SUMMARY			
NO LESION REPORTED	3	2	3
ANIMAL MISSING/NO NECROPSY			1
AUTO/NECROPSY/HISTO PERF	3	2	
AUTO/NECROPSY/NO HISTO		1	
AUTOLYSIS/NO NECROPSY	1		3

NUMBER OF ANIMALS WITH TISSUE EXAMINED MICROSCOPICALLY
* NUMBER OF ANIMALS NECROPSIED

Review of the Bioassay of 1,5-Naphthalenediamine* for Carcinogenicity by the Data Evaluation/Risk Assessment Subgroup of the Clearinghouse on Environmental Carcinogens

June 29, 1978

The Clearinghouse on Environmental Carcinogens was established in May, 1976, in compliance with DHEW Committee Regulations and the Provisions of the Federal Advisory Committee Act. The purpose of the Clearinghouse is to advise the Director of the National Cancer Institute (NCI) on its bioassay program to identify and to evaluate chemical carcinogens in the environment to which humans may be exposed. The members of the Clearinghouse have been drawn from academia, industry, organized labor, public interest groups, State health officials, and quasi-public health and research organizations. Members have been selected on the basis of their experience in carcinogenesis or related fields and, collectively, provide expertise in chemistry, biochemistry, biostatistics, toxicology, pathology, and epidemiology. Representatives of various Governmental agencies participate as ad hoc members. The Data Evaluation/Risk Assessment Subgroup of the Clearinghouse is charged with the responsibility of providing a peer review of reports prepared on NCI-sponsored bioassays of chemicals studied for carcinogenicity. It is in this context that the below critique is given on the bioassay of 1,5-Naphthalenediamine for carcinogenicity.

The reviewer agreed with the conclusion in the report that 1,5-Naphthalenediamine was carcinogenic in treated female rats and in both sexes of mice. He noted that the study was conducted in a room in which other compounds were under test. Based on the experimental findings, he concluded that 1,5-Naphthalenediamine may pose a carcinogenic risk to humans. The reviewer moved that the report on the bioassay of 1,5-Naphthalenediamine be accepted as written. The motion was approved without objection.

Clearinghouse Members present:

Arnold L. Brown (Chairman), Mayo Clinic
Paul Nettesheim, National Institute of Environmental Health Sciences
Verne Ray, Pfizer Medical Research Laboratory
Verald K. Rowe, Dow Chemical U.S.A.
Michael B. Shimkin, University of California at San Diego
Louise Strong, University of Texas Health Sciences Center

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* Subsequent to this review, changes may have been made in the bioassay report either as a result of the review or other reasons. Thus, certain comments and criticisms reflected in the review may no longer be appropriate.

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